ABSTRACT

Title of Document: MANAGEMENT AND CONSERVATION OF BROOK TROUT IN WESTERN MARYLAND

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Widespread declines have been observed in the abundance, distribution, and size structure of Brook Trout for nearly 200 years. Although broadly distributed, Brook Trout are very sensitive to environmental disturbances, and populations continue to disappear. Environmental change further threatens the persistence of wild Brook Trout, and even currently secure populations may be at risk. Life history variation and population substructures further confound management, and their potential influences on population dynamics warrant further investigation.

The objectives of my dissertation were to characterize Brook Trout populations in western Maryland and use this information to forecast alternative futures. We used a large-scale mark-recapture survey (>3,000 marked fish), molecular tools, and simulation modeling to gain a comprehensive understanding of the structure and function of Brook Trout populations in western Maryland.

We found that rapid visual assessment was a valid technique (92% accuracy after training) for determining sex in Brook Trout. We found significant variability in individual growth rates (0-144 $mm \cdot y^{-1}$), with marked influences of year, sex, size, and stream. We also detected the presence of cryptic metapopulations occurring on a small spatial scale and in the absence of physical barriers to movement. Population substructures such as sex or lineage are easily overlooked, yet they may have measurable and potentially important differences in vital rates.

Simulation modeling under current and alternative conditions suggested that environmental stochasticity exerts a strong influence on the population dynamics of wild Brook Trout in western Maryland. Population dynamics were driven by pulse-driven recruitment that was weakly related to spawner abundance. Changes in adult survival, representative of a range of management scenarios, had a considerable impact on population resilience. Conversely, changes in the growth rates of Brook Trout resulted in small changes to population resilience. Enhanced adult survival resulted in a greater abundance of large fish. Collectively, these results suggest regulatory approaches may offer some utility in promoting population resilience while enhancing the quality of the fishery, but are likely insufficient to fully offset the impacts of predicted environmental changes.

MANAGEMENT AND CONSERVATION OF BROOK TROUT IN WESTERN MARYLAND

By

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Dissertation submitted to the Faculty of the Graduate School of the
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Dedication

This work is dedicated to Caitie ‘Chomes’ Homan (1988-2013). Caitie was a great friend, passionate about brook trout, and selflessly volunteered to help with my research.

Acknowledgements

I would like to thank my advisor, Bob Hilderbrand, for providing excellent mentoring and ample resources for me to succeed, while allowing me the freedom to pursue additional research projects alongside my dissertation. My graduate committee (Ray Morgan, Michael Wilberg, Tim King, and Curry Woods) provided thoughtful feedback to my study design and helped me to maximize my potential as a fisheries scientist.

The scale and scope of my dissertation research would never have been possible without the generous support of the Maryland Department of Natural Resources Fisheries Service. Specifically, I would like to thank Matt Sell, Alan Heft, and the Mt. Nebo Fisheries office for their hard work during the seven electrofishing samples conducted for my dissertation. They conducted over 74 stream kilometers of survey work during the course of this study – truly an impressive effort. Additionally, the DNR Fisheries Service provided the PIT tags used in the study and helped with the construction and installation of the stationary PIT antenna arrays.

I would like to specially thank Steve Keller and Tim King for opening my eyes to the impressive utility of genetics in fisheries management, and for patiently teaching me how to use it successfully.

Many others assisted me in the completion of this dissertation. Each core research chapter (2-5) contains a list of specific acknowledgements. I am deeply appreciative of every person who helped bring my dissertation to fruition. Thank you very much!

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Chapter 1: Introduction

Background and Project Context

Brook Trout (*Salvelinus fontinalis*) are native to the eastern United States, where they are top predators in many coldwater streams. Historically, wild Brook Trout populations supported a considerable fishery, with anglers sometimes reporting catches of dozens of large trout on a single outing (Smith 1833, Karas 1997). Although the nature of the fishery has changed, Brook Trout remain culturally, economically, and ecologically important, as large numbers of recreational anglers target wild trout across their native range, contributing millions of dollars of economic benefits to rural communities (Karas 1997, Greene et al. 2005).

Widespread declines have been observed in the abundance, distribution, and size structure of Brook Trout for nearly 200 years (Smith 1833, Hudy et al. 2005, Heft 2006). Although broadly distributed, Brook Trout are very sensitive to environmental disturbances, and populations continue to disappear. In response, Brook Trout are the focus of substantial conservation efforts by private non-profit organizations as well as local, state, and federal agencies across their native range.

Brook Trout are the only salmonid native to Maryland. Over the last 100 years, this species has declined precipitously and it is now largely restricted to isolated headwater populations (Hudy et al. 2005, Heft 2006; Figure 1). In 2006, the Maryland Department of Natural Resources released their Maryland Brook Trout Management Plan, identifying anthropogenic impacts to Brook Trout including land use change, climate change, and population fragmentation (Heft 2006). The impacts of each of these stressors are expected to continue or worsen, leaving an uncertain future for Brook Trout

in Maryland and a clear need for strategic management. At the outset of my graduate research, five consecutive years of poor recruitment on the upper Savage River (Figure 2) were cause for immediate concern.

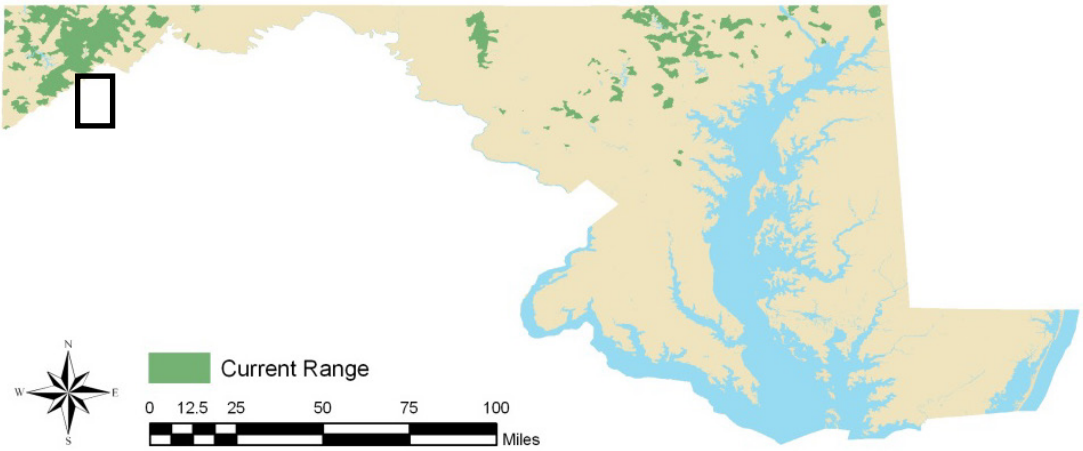

Figure 1. Present distribution of Brook Trout in Maryland based on records obtained from the Maryland Department of Natural Resources Fisheries Service. Study area is indicated by the black box.

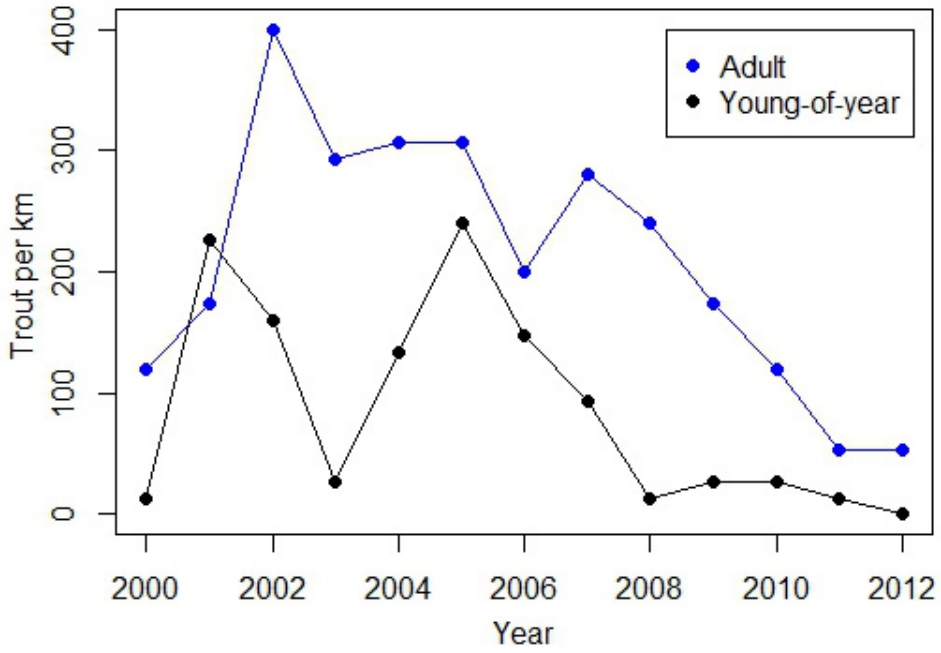

Figure 2. Density of adult and young-of-the-year Brook Trout on the upper Savage River mainstem (2000-2012). Data included in this figure were provided by the Maryland Department of Natural Resources Monitoring and Non-tidal Assessment Division.

Environmental change

Human activities are causing profound environmental change (Fox 2007), to the extent that we have entered a new geologic epoch (Zalasiewicz et al. 2008). Among the changes, climate change has been identified as one of the most serious threats to the biosphere in our era, and its effects will be especially pronounced on species living on the edge of their thermal tolerance, such as Brook Trout in the Mid-Atlantic region (Meisner 1990, Ries and Perry 1995, Eaton and Scheller 1996, Parmesan and Yohe 2003). In the northeast, mean annual temperatures have increased by 2°C since 1970 and are forecast to increase by an additional several degrees over the next few decades (Karl et al. 2009). Furthermore, the frequency of extreme weather events is increasing – a trend that is expected to continue or worsen (US EPA 1998, IPCC 2001, NOAA 2008). Conversion of open space to developed land is a widespread phenomenon and has similar effects on streams – increased mean temperature and frequency of disturbance (Stranko et al. 2008, Poff et al. 2006).

Temperature increases and extreme hydrologic events have pronounced effects on aquatic organisms (Palmer et al. 1992, Montgomery et al. 1999, Lake 2000, Fausch et al. 2001, McMahon et al. 2007). In Brook Trout, these conditions can cause acute mortality and or deterministic declines by changing key vital rates within a population (Ries and Perry 1995, Hakala and Hartman 2004, Roghair et al. 2002, Robinson et al. 2010). Predicted changes in the frequency and magnitude of acute disturbance coupled with a

chronic temperature increase may nonlinearly alter the ability of a species to recover from deleterious events, making even currently secure populations at risk.

Management to promote resilience has been suggested as a means to mitigate the effects of environmental change on natural resources (Hughes et al. 2003, Millar et al. 2007). In the context of fisheries, managers may adjust fishing regulations or improve habitat to offset changes in vital rates, but without an understanding of the range expected outcomes, these regulations are a shot in the dark.

Forecasting population responses to change

Forecasting population responses to global change can be a very powerful management tool for species of conservation concern such as the eastern Brook Trout (*Salvelinus fontinalis*). To understand the effects of environmental change and management activities, we can use models incorporating current and expected changes in vital rates to forecast population level responses (Hilderbrand et al. 2007). This type of approach allows us to test hypotheses using factors that are not experimentally tractable, but requires an understanding of the structure and function of the population of interest.

Brook Trout exhibit considerable life history variability, especially with respect to growth and longevity, but also in other key life history traits, such as movement, survival and age-at-maturity (Power 1980, Kazyak 2011). Such plasticity likely exists between the sexes and lineages (Gross 1996, Hutchings and Gerber 2002, Curry 2005, Chernoff and Curry 2007), but model parameters tend to homogenize across groups. If substructure-specific data changes our inferences on population dynamics, we may be currently mismanaging our aquatic resources without even realizing it. Capturing this intra-

population variation is important to adequately understand and manage population responses to environmental change, yet this approach is in its infancy (Frank et al. 2011).

Objectives

The overarching goal of my research is to forecast alternative futures for Brook Trout populations in Western Maryland, operating under the basic premise that key vital rates are impacted by environmental change and management strategies.

I will address the following key questions:

- Q_1. What management strategies are best suited to enhancing western Maryland Brook Trout populations?
- Q_2. In what ways can we expect predicted environmental changes to impact Brook Trout population dynamics? How can we manage Brook Trout to mitigate the impacts of environmental change?
- Q_3. How do vital rates vary among population substructures?

Through this dissertation, it is my intent to address key management needs for the Maryland Department of Natural Resources while advancing fisheries science.

Study Area

In Maryland, Brook Trout populations are clustered in three areas, roughly corresponding to the Piedmont, Catoctin Mountains, and Appalachian Plateau (Figure 1). I will focus on Brook Trout on the Appalachian Plateau, where populations are most robust and are the most likely to persist into the future based on climate change projections (Meisner 1990).

Within the Appalachian Plateau, the Savage River watershed hosts relatively robust Brook Trout populations and represents a regionally important stronghold for Brook Trout based on fish density and the large, interconnected network of suitable streams. A large portion of this watershed has been placed under special angling regulations, prohibiting the use of live bait or harvest of Brook Trout. This area offers an excellent opportunity to follow large numbers of Brook Trout through space and time while evaluating the effectiveness of the special regulations.

For the purposes of my dissertation research, I will focus on two study areas within the Savage River watershed – Big Run and Middle Fork (Figures 3-5). Both study watersheds are almost entirely on public land, and road and trail networks afford access to the study sites. Thus, these streams are ideal locations to study the structure and function of a population in a relatively undisturbed setting. The aquatic habitats are representative of many Brook Trout streams in the eastern portion of the species range. The Maryland Department of Natural Resources has also monitored these populations for several decades, offering insight into long-term population trends. Although the streams are in relatively intact condition, both were historically stocked with Brook Trout from Bear Creek Hatchery (Heft 2006) and their watersheds logged over 100 years ago. This

history is consistent with the stocking and land use records of many other brook trout streams in our region.

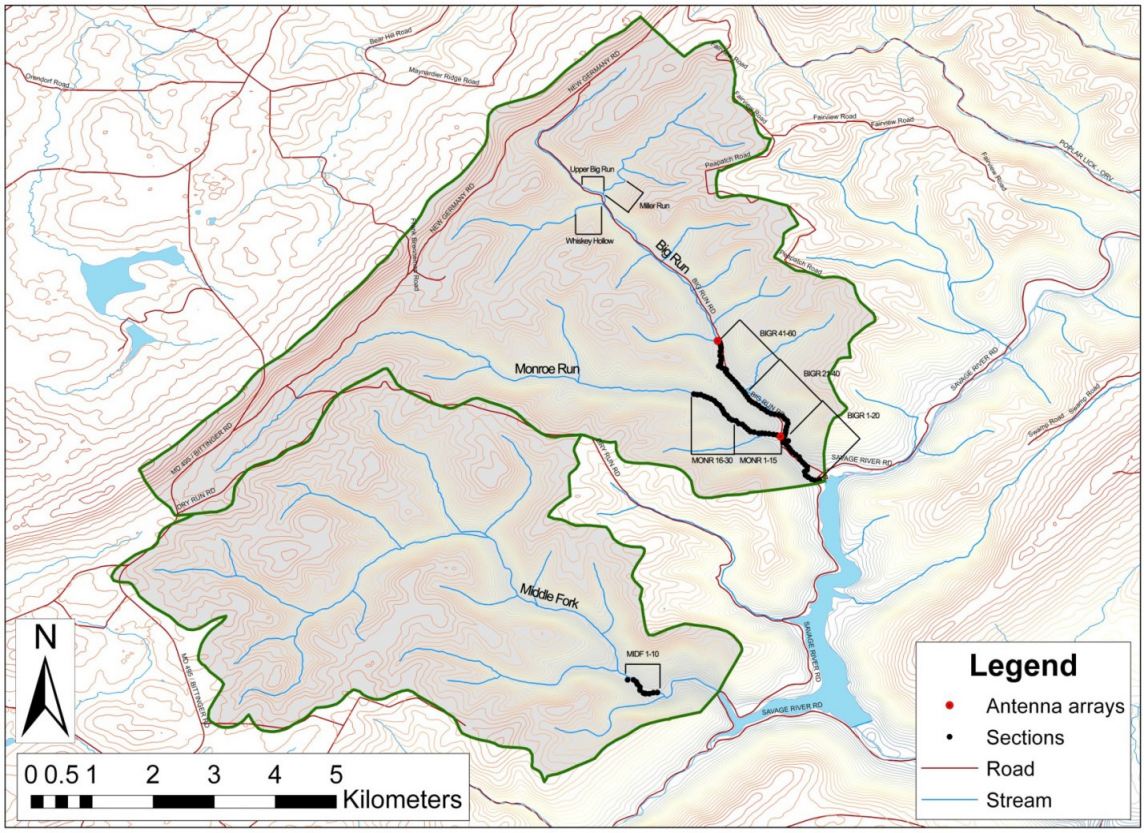

Figure 3. Core research areas within the Savage River watershed. Contour lines represent 50 ft intervals.

Figure 4. Big Run during summer low flow conditions.

Figure 5. Middle Fork of the Savage River near the study area at moderate flow.

Within Big Run, the study area consists of 4.5 km of stream delineated into 90 contiguous 50 m study sections. The study area starts at the confluence of Big Run and Savage Reservoir and continues upstream for about 1 km to the confluence of Big Run and Monroe Run. From here, our study area continues upstream for an additional 2 km on Big Run and 1.5 km on Monroe Run. The Big Run drainage supports a typical Appalachian Plateau stream community, dominated by Brook Trout, Blue Ridge Sculpin (*Cottus caeruleomentum*), and Eastern Blacknose Dace (*Rhinichthys atratulus*) as well as at least five other fish taxa (Table 1). During typical flow conditions, Big Run is generally 2-6 m wide with extensive shallow cobble habitats and some deeper pools.

Middle Fork, another tributary flowing directly into Savage Reservoir, will serve as a complimentary study area to boost our spatial coverage in the watershed and allow for comparisons among tributaries. On this stream, our study area focuses on a 0.5 km reach broken into 10 contiguous 50 m sections. The fish community in Middle Fork is very similar to that of Big Run, with the addition of an occasional White Sucker (*Catostomous commersoni*; Table 1). This portion of our study area is considerably more difficult for anglers to access, requiring a walk in excess of 1 km to reach the sites. Habitat conditions are similar between Middle Fork and Big Run, although Middle Fork is a slightly larger stream (3-7 m wide under typical conditions) with more deep pools.

Table 1. Relative abundance of fish taxa encountered during biannual electrofishing surveys at the two study areas within the Savage River watershed, Maryland.

			Relative Abundance	
Family	Scientific Name	Common Name	Big Run	Middle Fork
Catostomidae	*Catostomus commersoni*	White Sucker	-	Rare
Cyprinidae	*Rhinichthys cataractae*	Longnose Dace	Common	Common
	Rhinichthys atratulus	Eastern Blacknose Dace	Abundant	Abundant
Centrachidae	*Ambloplites rupestris*	Rock Bass	Rare	Rare
Cottidae	*Cottus caeruleomentum*	Blue Ridge Sculpin	Abundant	Abundant
	Cottus girardi	Potomac Sculpin	Rare	Rare
Percidae	*Etheostoma flabellare*	Fantail Darter	Common	Common
Salmonidae	*Salvelinus fontinalis*	Brook Trout	Abundant	Abundant
	Oncorhynchus mykiss	Rainbow Trout	Rare	Rare

Within the study area, forecast environmental changes may impact Brook Trout populations. Karl et al. (2009) reports that mean temperatures have increased 2°F since 1970 in the northeastern United States, with further increases of 2.5-4°F and 1.5-3.5°F expected in the winter and summer, respectively (Figure 6). Additionally, these authors describe a suite of other projected impacts, including an increased frequency of extreme heat, drought, and extreme precipitation events. Furthermore, it is anticipated that ice on streams will breakup earlier in the year, snowpack will decrease, and forest communities will change. Local changes in forest cover and water withdrawals, such as those that sometimes occur in conjunction with Marcellus shale gas drilling, may also change streams in our study area (Eshleman and Elmore 2013). All of these projected changes in the environment have the potential to alter the vital rates of Brook Trout in the Savage River watershed, but due to the complex nature of the expected environmental changes, it is impossible to say for certain how vital rates will be affected.

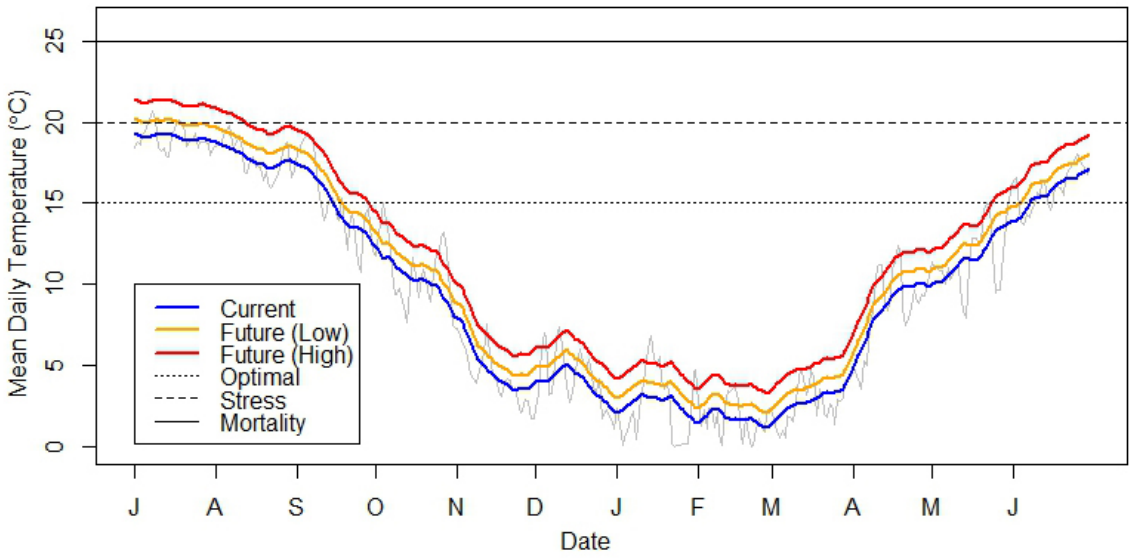

Figure 6. Sample temperature profiles for Big Run under current conditions and warming scenarios. Data shown represents mean daily temperatures (smoothed using a 21 d moving average) derived from a temperature logger deployed at Savage River Road from July 2012-June 2012. Warming scenarios are based on the projected air temperature increases of Karl et al. (2009) and unpublished work by Bob Hilderbrand on the relationship between air and water temperatures in Maryland streams.

Chapter 2: Rapid visual assessment to determine sex in Brook Trout

(At the time of dissertation submission, this chapter was published in volume 33 in the North American Journal of Fisheries Management, titled "Rapid visual assessment to determine sex in Brook Trout" by D.C. Kazyak, R.H. Hilderbrand, and A.E. Holloway; pages 665-668. As the lead author, I contributed substantially to the design and implementation of this study and therefore have included it as a chapter in my dissertation.)

Abstract

Although sex-specific processes play a considerable role in the ecology of many fishes, nonlethal tools to determine sex in most species outside of the spawning season are lacking. We identified a suite of sexual characteristics in Brook Trout *Salvelinus fontinalis* by surveying the available literature, consulting biologists, and reviewing images of known-sex individuals. Using pre-training and post-training testing of fisheries professionals, we assessed the utility of color and morphology for sexing fish across a length gradient (110–251 mm TL) by presenting images of mature fish collected during the spawning season. Rapid visual assessment proved to be an effective approach for determining the sex of Brook Trout. Average accuracy significantly improved from 71.5% before training to 92% after training, and the proportion of fish scored as unknown was reduced (from 9.5% before to 2.0% after training). We found a small yet significant positive relationship between TL and the proportion of fish that were correctly sexed. With the exception of a single 113-mm male, all individuals were correctly sexed by at least 79% of respondents after training. No significant differences were found among respondents based on education, experience, or confidence level. The effectiveness of rapid visual assessment also did not significantly differ between male and female Brook

Trout. Rapid visual assessment is a viable technique for the determination of sex in Brook Trout.

Introduction

Practical methods of sex determination in fishes are rare but need to be made available to researchers and managers alike. Ideal techniques are nonlethal, quick, and affordable while also allowing sex determination throughout the year and at all stages of ontogeny. Such methods open new doors in fisheries research to identify sex-specific vital rates and their implications for population-level processes; for example, Brook Trout *Salvelinus fontinalis* exhibit considerable life history variation, and tactics may vary by sex (Hutchings et al. 1999; Hutchings 2006). An enhanced understanding of sex-specific life history variability may improve our ability to manage fishes, but candidate approaches must be evaluated before they can be used.

Male and female Brook Trout are known to be genetically (Phillips et al. 2002) and morphologically distinct (Power 1980), but there are no established, practical methods to determine the sex of Brook Trout during periods outside of the spawning season. A useful approach would be to use known-sex individuals obtained during the spawning season to identify characters that should be retained throughout the year. Our objectives were to (1) evaluate the effectiveness of rapid visual assessment (RVA) for sexing Brook Trout; (2) determine whether the effectiveness of RVA can be improved through a short training session; and (3) examine factors that contribute to the effectiveness of RVA for determining the sex of Brook Trout.

Methods

We used backpack electrofishing to collect 111 Brook Trout from Big Run, a tributary to the Savage River in western Maryland. Fish were collected on 6–8 October 2011, approximately one week prior to the observed peak of spawning activity. This sampling period was chosen to maximize the proportion of mature, known-sex individuals observed within the population. Each fish was mildly anesthetized using tricaine methanesulfonate (80 mg/L; buffered with 0.2-mM $NaHCO_3$, pH= 7).We measured the TL (mm) of each fish and attempted to manually express gametes from all captured individuals. If gametes were expressed, the fish was identified as male (n = 36; 113–243 mm TL) or female (n = 29; 110–251 mm TL) accordingly. The fish were collected in a no-kill management zone and could not be lethally sampled. Consequently, the sex of individuals that did not express gametes was considered to be unknown (n = 46; 107–216 mm TL), and these individuals were excluded from the study. Standardized photographs of the left side of each individual were collected by using consumer-grade digital photography equipment mounted on a tripod at a fixed distance (20 cm) above a white background. We analyzed these photographs, reviewed pertinent literature (Power 1980; Holloway 2012), and conducted informal conversations with biologists to identify secondary sexual characteristics in Brook Trout.

At the East Coast Trout Management and Culture Workshop V in 2012, we conducted a survey of 57 voluntary participants (University of Maryland Institutional Review Board Project 329909-1). The survey was broken into four components. First, we collected demographic data from each participant (education level, experience, and confidence in sex identification). Next, participants were shown a series of slides (hereafter, pre-training test) developed from a randomly selected subset (n = 25) of our

images of known-sex fish. Each slide was shown for 10 s and clearly stated the TL of the fish. For each slide, participants were asked to identify the sex of the fish as male, female, or unknown. Based on the results of a morphological analysis of sexual dimorphism in Brook Trout (Holloway 2012) and a literature review, we presented a 5-min training session highlighting a suite of secondary sexual characteristics that are useful for distinguishing male and female Brook Trout (Table 2; Figure 7). Also included in the training were pictures of additional known-sex fish, and distinguishing characters were highlighted. Finally, the pre-training images were shown to participants in a random order (hereafter, post-training test), and participants were once again asked to identify each fish as male, female, or unknown.

A paired *t*-test was used to compare the pre-training and post-training percentages of fish that were correctly identified or that were identified as unknown. We used a two-sample *t*-test to compare the accuracy of RVA for males and females, and ANCOVA was used to evaluate changes in the accuracy of RVA before and after training and with respect to fish TL. Analysis of variance was used to determine whether significant differences in accuracy existed among demographic attributes of the survey participants. Because the response data were percentages, we used an arcsine transformation for all analyses to better meet the assumptions required for *t*-tests, ANOVA, and ANCOVA. Finally, we used linear regression on untransformed data to evaluate whether pre-training performance could predict post-training improvement.

Table 2. Characters used to distinguish male and female Brook Trout.

Male	Female
Body slab-sided†	Body fusiform
Deeper head	Head not as tall
More colorful†	Less colorful
Longer, pointed snout	Shorter, rounded snout
Kype present (on larger fish) †	Kype absent
Black smudges on sides on ventral surface†	Black smudges absent or less bold

† First reported by Power (1980).

Figure 7. Comparison of external features used in rapid visual assessment of sex in Brook Trout. Note the pointed snout, slab-sided body, and dark black abdominal smudge on the male (top) compared to the rounded snout, more fusiform body, and the relative lack of the black abdominal smudge on the female (bottom). These individuals were collected during the October spawning season.

Results

A suite of secondary sexual characteristics was identified in Brook Trout (Table 2; Figure 7). In general, males were laterally compressed, with a deep head and a long, pointed snout. In contrast, females were torpedo-shaped, with a smaller head and a short, rounded snout. On large males, a kype was sometimes present. Males tended to be more brightly colored than females, although vibrant females were also observed. Many males had dark smudges along the sides of the abdomen. These smudges were less common and typically less pronounced on female Brook Trout. While none of these features was strictly dichotomous, the suite of characters taken together appeared to offer considerable insight into the sex of an individual. Anecdotally, the black smudges are retained throughout the year but are less pronounced at times other than the spawning season.

Survey participants were able to correctly identify the sex of Brook Trout with much greater accuracy than simple guessing, both before the training session (mean = 71.5%, SD = 16.0%; $t_{56} = 9.8$, $P < 0.001$) and after training (mean = 92.0%, SD = 5.7%; $t_{56} = 32.2$, $P < 0.001$). However, the percentage of fish that were correctly sexed was significantly greater after the training session ($t_{56} = -11.3$, $P < 0.001$). In addition, the percentage of fish that were identified as being of unknown sex was significantly lower after the training session (mean = 2.0%, SD = 3.5%) than before training (mean = 9.5%, SD = 10.1%; $t_{56} = 7.7$, $P < 0.001$).

There was no evidence of an interaction between fish TL and training with regards to the effectiveness of RVA. Length had a weak positive relationship with the percentage of fish that were correctly sexed for both pre-training and post-training tests ($R^2 = 0.14$, $P < 0.05$; Figure 8). However, with the exception of a single 113-mm male, all individuals were correctly sexed by at least 79% of respondents after training (Figure

8). When that individual was omitted from our analysis, the average post-training accuracy was 94.4%. There was a strong negative relationship between the pre-training scores and the improvement that occurred after training ($n = 57$, adjusted $R^2 = 0.87$, $P < 0.001$; Figure 9): respondents that scored poorly before training exhibited significantly greater post-training improvement than respondents that had high scores on the pre-training test. Accuracy during the pre-training and post-training tests was not affected by the respondents' experience, position, education, or perceived confidence in sexing fish ($P > 0.05$). There was no difference in the accuracy of sexing male fish versus female fish ($t_{22, 28} = -1.7$, $P = 0.10$).

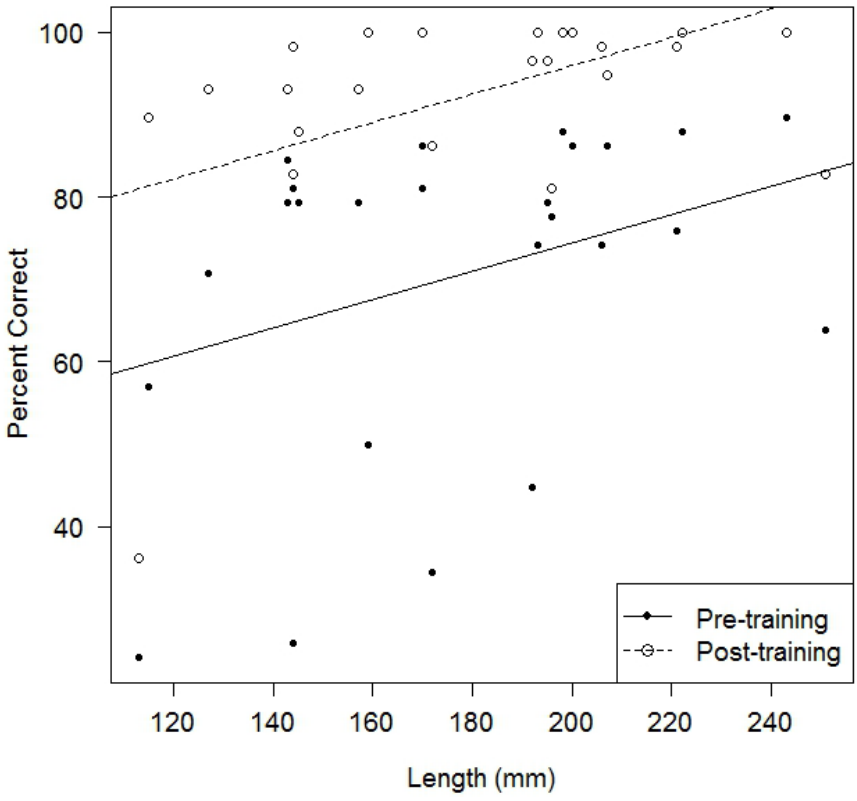

Figure 8. Training and fish length exhibit a significant positive relationship with the percentage of Brook Trout correctly sexed using rapid visual assessment. Untransformed data shown for visual clarity. Note the substantially reduced variation in the post-training scores compared to the pre-training scores.

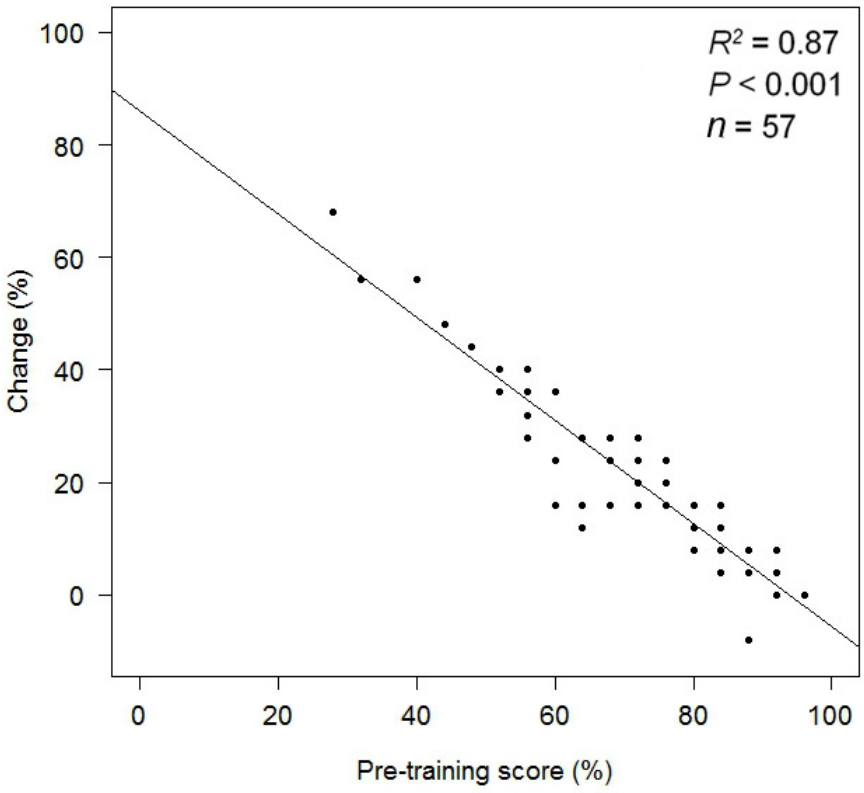

Figure 9. Linear regression showing the relationship between pre-training scores and post-training improvement. Pre-training scores showed a strong inverse relationship with improvement following a short training session.

Discussion

Rapid visual assessment based on morphology has been successfully used to determine the sex of Pacific salmon *Oncorhynchus* spp. (Beacham and Murray 1986; Brykov et al. 2010). Our study demonstrates the effectiveness of RVA for use with mature, riverine Brook Trout during the spawning season. Accuracy rates were high (mean = 92.0%) and uncertainty was low (mean = 2.0%) after training had been provided. After the smallest individual fish in the survey was excluded from analysis, overall accuracy rates exceeded 94%. Sexual maturity in small Brook Trout (<120 mm) is uncommon but well documented in stream-dwelling populations (McFadden 1961;

Wydoski and Cooper 1966). In other locations, where larger Brook Trout are the focus of field studies, we expect that secondary sexual characteristics will be more pronounced and that RVA will be even more effective. Although some participants had very high scores (up to 96% accuracy) on the pre-training test, marked overall improvement (mean = 20.5%) during the post-training test suggests that knowledge of a few key morphological characteristics can significantly improve a person's ability to accurately sex Brook Trout. There were no significant differences in performance based on demographic variables (e.g., experience, education level, or confidence) before or after training, further suggesting that this approach can be rapidly learned by fisheries personnel.

The RVA approach appears to perform well across sizes and sexes within the Big Run population of Brook Trout. Accuracy rates for male and female Brook Trout were not different, and TL differences, although statistically significant, did not have a strong impact on the accuracy of RVA. Larger fish were correctly sexed more often, probably due to their more pronounced secondary sexual characteristics. However, even the smaller fish were correctly sexed at least 79% of the time, which approaches the accuracy rates reported by Beacham and Murray (1986) for spawning Pacific salmon. Brook Trout are thought to exhibit morphological variation among populations, and transferability remains untested. Although we cannot directly evaluate the transferability of RVA, we are encouraged that in the pre-training test, some biologists from other regions effectively sexed Brook Trout collected at Big Run, thus indicating that secondary sexual characteristics are conserved between populations.

The Brook Trout used in this study were collected during the spawning season in order to allow definitive determination of sex. However, almost every morphological character that we used should be visible throughout the year. Coloration may vary with season, with males in particular showing bright oranges and reds. Another key, sexually dimorphic character is the shape of the snout. Males tend to have more angular snouts, whereas the snouts of females are rounded, and this difference was noticeable even in the smallest fish we were able to accurately sex (Holloway 2012). Exaggerated head features have been observed in males of other salmonids (Fleming and Gross 1994; Casselman and Schulte-Hostedde 2004), and these sexual characteristics appear to be retained after an individual has reached reproductive age (Janhunen et al. 2009). Survey participants also noted that around the spawning season, females often exhibit a swollen vent. This additional character may help biologists when using RVA in the field, but it was not feasible for RVA based on the photographs presented in our survey. Although the accuracy of RVA may vary to some degree, the primary characters used to determine sex in Brook Trout should be effective throughout the year.

Overall, RVA appears to be a viable approach for determining the sex of Brook Trout in some applications. This approach is fast, affordable, and minimally invasive, but it lacks the level of certainty that may be required for some applications and it may not be as effective for use with immature individuals. Identification of additional sex-specific traits that are effective at all stages of ontogeny and that have been cross validated with genetic markers would substantially enhance our ability to determine sex-specific life history attributes and variables for the purpose of better management of fishes.

Acknowledgments

Field discussions with Joe Zydlewski (U.S. Geological Survey, Maine Cooperative Fish and Wildlife Research Unit) inspired this research project. Mark Colaw and Wes Wagner aided with the field component of this study. Alan Heft and Matt Sell (Maryland Department of Natural Resources) facilitated the implementation of the survey. This paper is scientific series contribution number 4757 from the University of Maryland Center for Environmental Science, Appalachian Laboratory.

Chapter 3: Growth variation in a Mid-Atlantic Brook Trout population

(At the time of publication, this chapter was accepted for inclusion in Wild Trout XI: Looking Back and Moving Forward. As the lead author, I contributed substantially to the design and implementation of this study and therefore have included it as a chapter in my dissertation.)

Abstract

We used recapture data from >2,200 individually marked Brook Trout *Salvelinus fontinalis* to quantify their growth rates in three western Maryland streams. Individual growth rates varied greatly among fish, ranging from 0 to 144 mm·year^{-1}. More importantly, we found substantial growth differences among years, resulting in some fish in their second year being as long as four-year-old fish. Although hooking injuries and conspecific density were not important drivers of individual growth, we found sex-specific differences in growth, as males grew 10.5 mm·year^{-1} faster on average than females. We also found significant growth differences among reaches, highlighting the importance of downstream areas for Brook Trout growth. Strong inter-annual and sex-specific growth variation have major implications for population management, and have forced us to reassess our understanding of population dynamics because of the strong relationship between fish length and fecundity, our inability to confidently determine age from length-frequency plots, and the dominant, and largely uncontrollable, drivers of Brook Trout growth in these streams.

Introduction

Brook Trout *Salvelinus fontinalis* are a highly plastic and variable species whose life history, habitat use, movement patterns, morphology, and genetics may all vary markedly among and within populations (Power 1980; Perkins et al. 1993). In turn, growth and longevity in Brook Trout are also highly variable within and among

populations, resulting in considerable differences in population size structure and reproductive output (Hutchings 1993; Ficke et al. 2009). Brook Trout populations may be dominated by young and fast growing individuals, long-lived and slow growing fish, or represent an aggregation of individuals exhibiting disparate life history strategies (Dutil and Power 1980; Power 1980). Because size and reproductive output are closely linked (Hutchings 1993), growth rates have a strong influence on the dynamics of a population. If we can identify factors that are important controls of growth, we may be able to identify opportunities for managers to enhance wild trout populations.

Given the high variability observed in Brook Trout life history, local information is critical for effective management. The objectives of this study were to (1) describe growth variability in the Savage River watershed and (2) explore intrinsic, biotic, and abiotic factors that influence individual growth rates. We used observed growth at size to (3) calculate Von Bertalanffy growth trajectories for two years to highlight the importance of inter-annual variation of growth rates.

Methods

We conducted our study on two streams in the Savage River watershed of western Maryland (Figure 10). The Savage River watershed contains >100 km of interconnected coldwater streams and has been identified as a regionally important stronghold for wild Brook Trout. Within this drainage, our efforts focused on two tributaries: Big Run (4.5 km) and Middle Fork (0.5 km). Both streams are the focus of a large-scale tagging project and have been divided into contiguous 50-m study sections.

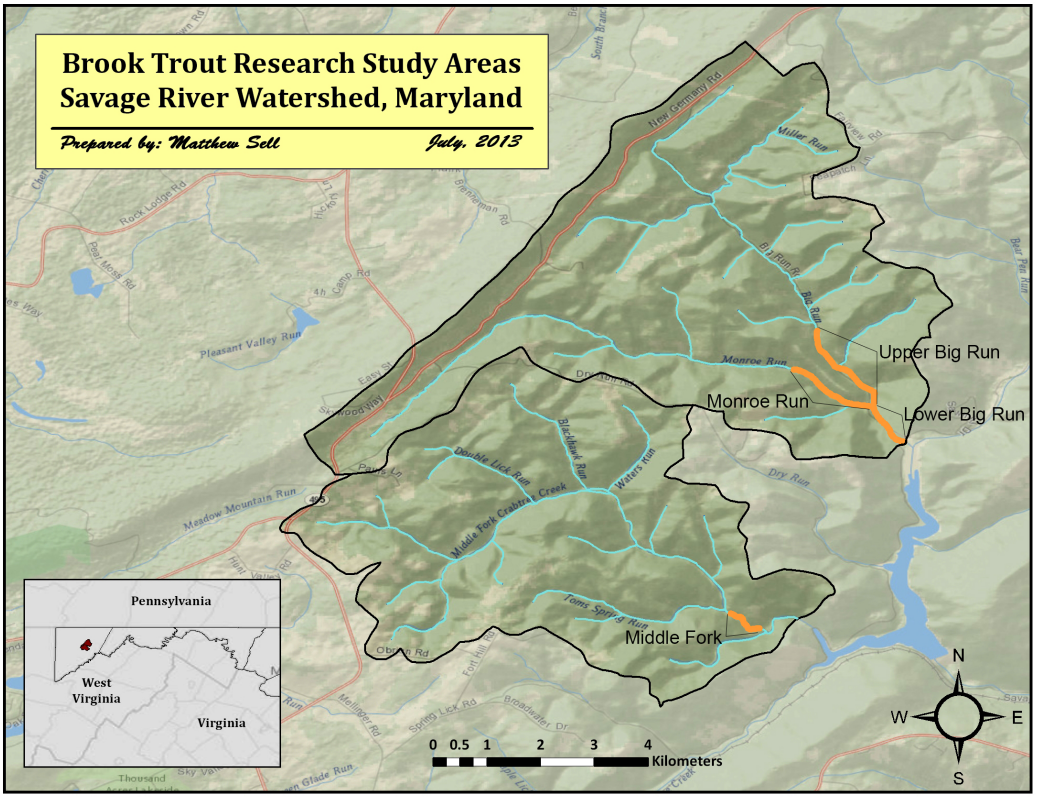

Figure 10. Location of study reaches with the Middle Fork and Big Run watersheds.

Fish were collected annually during early summer using three-pass backpack electrofishing surveys throughout each of the study sections. All fish were chemically anesthetized (80 mg·L^{-1} tricaine methanesulfonate buffered with 0.2 mM NaHCO3, pH = 7) prior to handling. Total length (mm) was measured and each individual was examined for hooking injuries. All individuals that had not been previously captured were surgically implanted with a 12-mm passive integrated transponder (PIT) tag (134.2 kHz ISO tag, Digital Angel Co., TX1411SST). Sex was determined for a subset of the tagged individuals during fall electrofishing samples conducted shortly before the peak of spawning. For all captured individuals, manual expression of gametes was attempted. For those individuals that did not express gametes, rapid visual assessment (Kazyak et al. 2013) was used to assign sex.

For the purposes of this study, we used absolute growth rate as our response variable (Equation 1). This metric of growth was selected because it is easy to interpret, scales linearly with individual length (Figure 11), and is directly compatible with the Fabens (1965) modification of the von Bertalanffy Growth model.

$$\text{Absolute growth} = \frac{L_1 - L_0}{t_1 - t_0} \qquad \text{(Equation 1)}$$

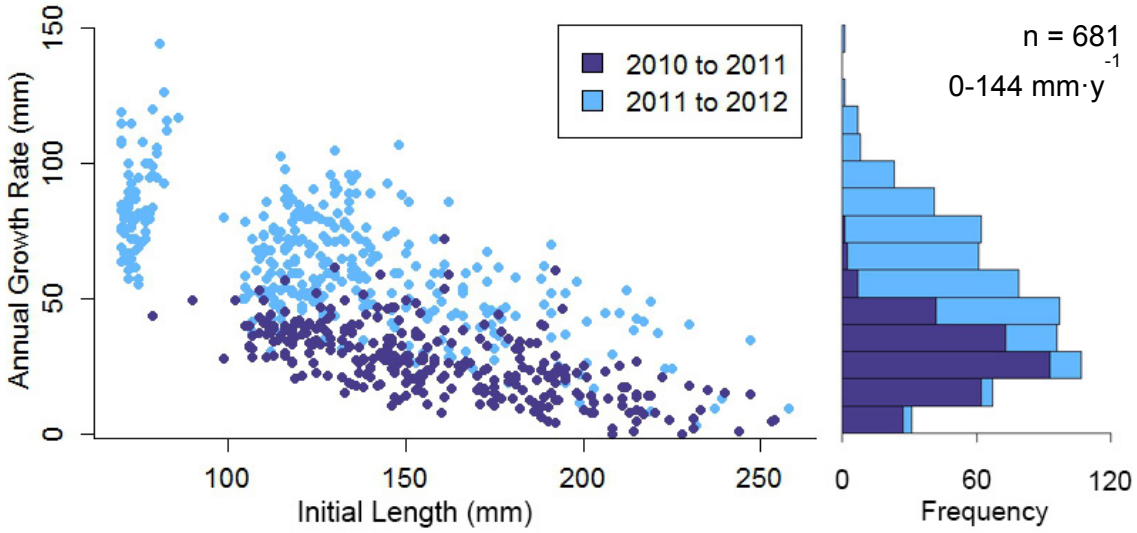

Figure 11. Observed annual growth rates versus initial length for two years. Growth rates generally slowed with individual size, but were markedly faster during the second year of the study.

To determine if conspecific density promotes spatial variability in growth rates, we derived Brook Trout biomass and adult counts from three-pass electrofishing surveys. Because our overall sampling efficiency was generally in excess of 95%, we used the abundance and biomass of our catch to represent Brook Trout density. The vast majority of recaptured individuals were found in the same section one year later. Consequently, we used an individual's location at the start of a growth interval to assign place-based covariates.

Using field data, we created a suite of *a priori* linear models using Program R (R Core Team 2012) to explain individual growth rates as a function of intrinsic, biotic, and abiotic factors (Table 3). Candidate variables included total length (mm), year, adult count, reach, sex, and hooking injuries. All predictor variables represent data collected at the start of the growth interval. Adult count equals the total number of adult Brook Trout (>100 mm TL) encountered in the study section at the start of the period at large. For the purposes of this analysis, the study area was divided into four reaches: Middle Fork, Monroe Run, Lower Big Run and Upper Big Run. We used the Akaike Information Criterion as a guide to identify top candidates for model selection. Among these models, we used the overall variance explained (R^2) and model complexity (k) to select a model which was best suited to elucidating key drivers of individual growth variation in our area.

We used the Fabens (1965) modification of the von Bertalanffy growth model to project size-at-age trajectories using data from each of the two years. Model outputs were used to calculate expected fecundity-at-age based on the length-fecundity relationship reported by Halfyard et al. (2008) for lotic Brook Trout, assuming fish mature in their second fall.

Table 3. Comparison of the structure and efficacy of a suite of *a priori* models developed to explain individual growth rates as a function of size, year, location, hooking injuries, sex, and competition. All place-based variables correspond to the section where an individual was captured at the start of the growth interval. The model favored by the authors is shown in bold. The number of estimated parameters is represented by *k*.

Model	Structure	*k*	AIC_C	ΔAIC_C	Adj. R^2
A	Length * Year + Adult Count * Year + Reach * Year + Sex	14	5203.58	0.00	82.4
B	Length * Year + Adult Count * Year + Reach * Year + Sex + Hooking Injury	15	5205.57	1.99	82.4
C	**Length * Year + Reach * Year + Sex**	**12**	**5210.20**	**6.62**	**82.2**
D	Length * Year + Reach * Year	10	5255.10	51.52	81.0
E	Length * Year + Sex	6	5492.09	288.51	72.9
F	Length * Year	4	5551.56	347.98	70.4
G	Length	2	5957.11	753.53	46.2

Results

We PIT tagged >2,200 individual Brook Trout during the summers of 2010 and 2011. These efforts yielded 681 recapture events during 2011 (21 male, 24 female, 267 unknown sex) and 2012 (66 male, 111 female, 198 unknown sex). The vast majority of recaptured individuals remained in the same section, although a few individuals made more extensive movements (Figure 12). Individual growth rates varied widely, ranging from 0 to144 mm·year^{-1} (Figure 11). Hooking injuries were detected in 3.2% of the tagged individuals that were subsequently recaptured and represented in the model.

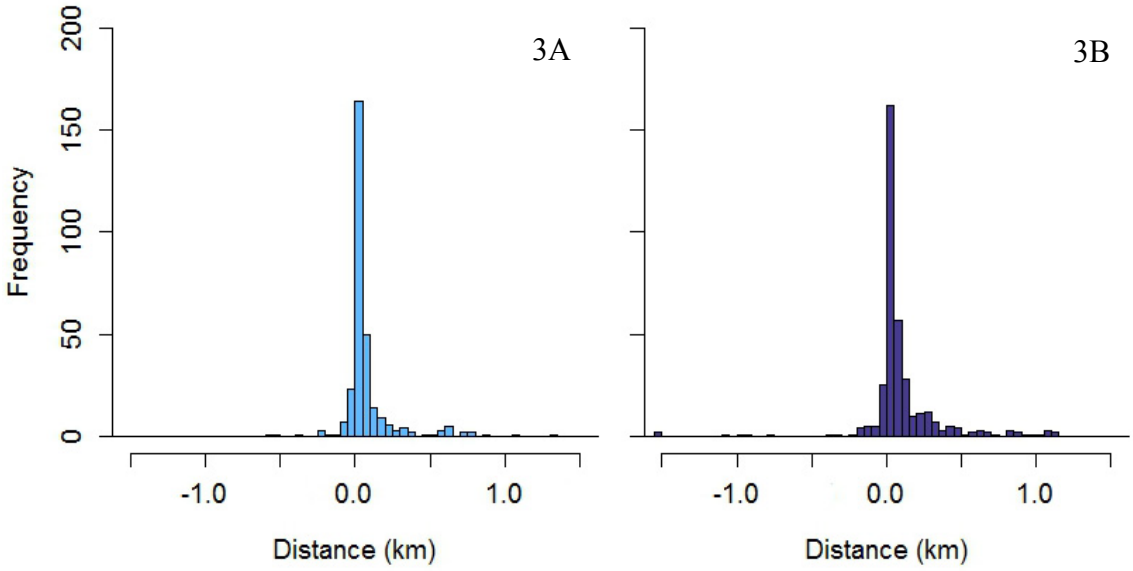

Figure 12. Histogram of observed distances between recaptures (June to June) for Brook Trout in the Savage River watershed, MD. Dispersal distances were similar between the first (3A) and second (3B) years of the study. Six outliers are omitted from plot 2B, and their dispersal distances were as follows: -1.55, 1.75, 1.90, 2.35, 2.65, and 2.70 km.

The best model (Model C; Table 3) explained 82% of the variation in growth (Figure 13) and was comprised of individual length and stream reach, the effects of which vary by year, and also sex. Growth rates were significantly lower in the first year ($\bar{x}$ = 26.57 mm·year^{-1}) relative to the second year ($\bar{x}$ = 63.32 mm·year^{-1}). This was reflected in the parameter estimates generated by our model, where individual growth rates varied considerably between the two years and to a lesser extent among stream reaches (Table 4). The negative effect of individual size on growth was also more pronounced during the second year (-0.40 mm·year^{-1}·mm^{-1}) compared to the first (-0.26 mm·year^{-1}·mm^{-1}; Table 4). Mean annual growth for male Brook Trout was 10.49 mm·year^{-1} greater than for females, while the growth rate for those fish where sex was unknown was intermediate (Table 4).

Von Bertalanffy growth trajectories observed during the first year of the study (2010 to 2011) resulted in much reduced size-at-age and fecundity-at-age when compared to projections using data from the second year (2011 to 2012; Figure 14). Based on the first growth interval, our modeled growth trajectory predicted an age-3 fish would be 182 mm and produce 280 eggs. For comparison, the rapid growth rates observed during the second year of the study yielded a predicted size of 246 mm for the same age, with females of this size expected to produce 545 eggs (Halfyard et al. 2008) – nearly double the predicted age-3 fecundity from the first year.

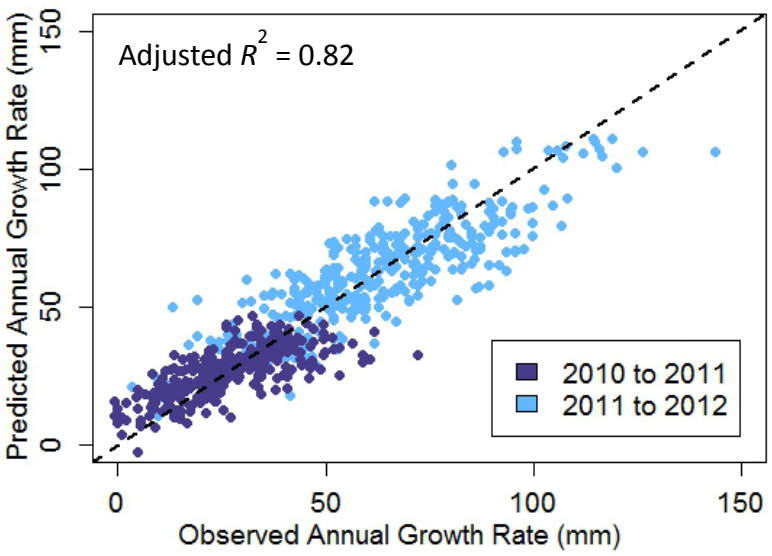

Figure 13. Observed versus predicted annual growth rates for individual Brook Trout. Predicted growth rates were derived using parameter estimates from model C.

Table 4. Estimated parameters derived from the selected model (C). All units are $mm \cdot y^{-1}$.

Variable	Observed Condition	Estimate ± SE	
		2010	2011
Reach	Monroe Run	62.43 ± 4.64	101.00 ± 10.23
	Lower Big Run	71.10 ± 4.94	128.33 ± 11.30
	Upper Big Run	63.75 ± 3.09	113.57 ± 6.70
	Middle Fork	65.93 ± 4.98	104.50 ± 10.57
Length	mm^{-1}	-0.26 ± 0.02	-0.40 ± 0.04
Sex	Female	0.00 ± 0.00	
	Male	10.68 ± 1.52	
	Unknown	3.75 ± 1.16	

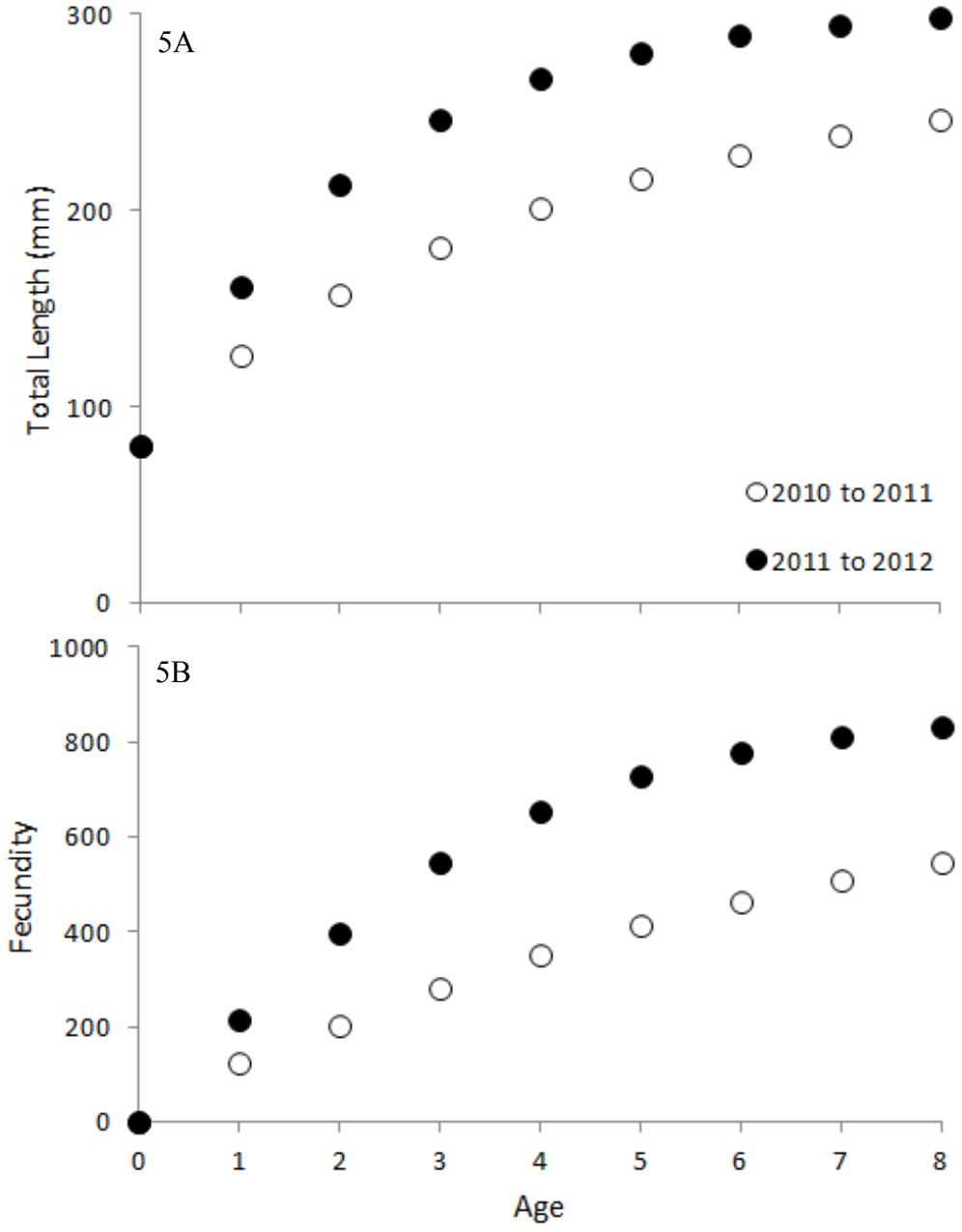

Figure 14. Mean length at age predictions based on the Von Bertalanffy growth model using data from two separate years (5A). Initial length at age-0 was fixed at 80 mm and reflects the typical length of new recruits during the electrofishing surveys. Expected fecundity based on projected growth trajectories and the length-fecundity relationship reported by Halfyard et al. (2008) for lotic Brook Trout, assuming fish mature in their second fall (5B).

Discussion

Our results show that size is a poor proxy for age, and consequently, we do not expect length-frequency histograms to provide accurate results if growth rate varies among years. Spatial, temporal, and sex-specific variation in growth rates are more than sufficient to confound the relationship between size and age. Differences among stream sections also suggest that some areas within riverine networks have greater Brook Trout growth than others. In both years, the highest growth rates were observed in the lower portion of Big Run. This is consistent with the idea that downstream reaches may be important feeding habitats for Brook Trout (Utz and Hartman 2006). Thus, management should strive to maintain these downstream, interconnected reaches because of their biological significance.

Hooking injuries did not influence individual growth rates, even though some fish had significant jaw deformities. Although simulations (Meka and Margraf 2007) have suggested there may be sublethal impacts of angling on salmonid growth, our field results are consistent with laboratory studies using live fish (Pope et al. 2007). However, our result should be viewed as preliminary, because our sample size of fish with hooking injuries was quite small.

The significantly lower growth of female Brook Trout has important implications for population dynamics and fisheries management. Since fecundity is tightly correlated with size in fishes (Blueweiss et al. 1978), slower female growth implies that actual reproductive output may be much lower than expected based on models lacking sex-specific data. We also expect males to dominate the largest size classes within the population, assuming equal life span between sexes. Future work may consider examining sex ratios across a range of size classes within a Brook Trout population.

Habitat quality has sometimes been associated with variation in growth rates in salmonids (Hayes et al. 1996). A preliminary examination of the direct effects of habitat quality did not show any strong signals, but missing data precluded a formal analysis. We used conspecific density as a surrogate for habitat quality and found it was not a useful predictor of individual growth rates despite density-dependent growth rates in other Brook Trout populations (Utz and Hartman 2006).

We documented clear differences in size- and fecundity-at-age trajectories between two years of the study. Additional years of data may help us understand the complete distribution of growth rates exhibited in our study populations. However, it is clear that population projections and management decisions based on a single year of data may be considerably different than those based on multiple years of data, which are required to capture the inherent variability within the population. Taken as a whole, our study highlights the importance of high-resolution, multi-year data when using science to inform management. Projected increases in environmental variability will only increase the importance of longer-term datasets.

Acknowledgements

The Maryland Department of Natural Resources was instrumental in providing financial and field support for this project. In particular, we would like to thank Alan Heft, Alan Klotz, and the Mt. Nebo fisheries team for their help with electrofishing and tagging. Numerous other volunteers assisted in the field, and for that we are grateful. Bob Gardner, Matt Fitzpatrick, Katia Engelhardt, Miriam Johnston, and Steve Keller provided useful feedback on the modeling approach used in this manuscript. We would also like to thank Michael Wilberg for his valuable insight on the analysis of this data.

Chapter 4: Hiding in plain sight: a case for cryptic metapopulations in Brook Trout

(This chapter is being prepared for submission as a peer-reviewed article by D.C. Kazyak, R.H. Hilderbrand, T.L. King, S.R. Keller, and V.E. Chhatre. As the lead author, I contributed substantially to the design and implementation of this study and therefore have included it as a chapter in my dissertation.)

Abstract

A fundamental issue in the management and conservation of species is how to define a population. Spatially contiguous fish populations occupying a stream network have often been considered to represent a population. While these fish may represent a single, homogenous population, there are several alternative scenarios that need to be considered. Conceptually, fish in spatially contiguous habitat may also represent multiple discrete populations, a single population with genetic isolation-by-distance, or a metapopulation. We used microsatellite DNA and a large-scale mark-recapture study to assess Brook Trout population structure. We found evidence for limited genetic exchange across small spatial scales and in the absence of barriers to physical movement. Mark-recapture and stationary PIT antenna records demonstrated that fish from two tributaries very seldom moved into the opposite tributary, but movements between the tributaries and mainstem were more common. Using Bayesian genetic clustering, we identified two genetic groups that exhibited significantly different growth rates over three years of study, yet survival rates were very similar. Our study highlights the importance of considering the possibility of multiple genetically distinct populations occurring within spatially contiguous habitats, and suggest the existence of a cryptic metapopulation: a spatially continuous distribution of organisms exhibiting metapopulation-like behaviors.

Introduction

The management and conservation of genetically distinct groups of organisms is a central pillar of modern conservation biology. However, defining populations can be an important challenge for understanding basic natural history as well as for recognizing appropriate units of biodiversity for conservation and management. Many populations show local adaptation to their environment, which may be expressed as variation among populations in life history strategies (Hutchings 1993), physiology (Eliason et al. 2011), and morphology (Quinn et al. 2001). Genetic differences among nearby populations may reflect a history of local adaptation, and provides a reservoir for potential adaptation to changing conditions in the future.

In order to manage populations, we need to know how they are structured. Inventories of genetic diversity and population size may be very misleading if population structures are not adequately understood (Chikhi et al. 2010). Similarly, groups of populations can exhibit a portfolio effect, thus increasing overall resilience and masking the dynamics of individual populations (Schindler et al. 2010). In both scenarios, our understanding of the status and trends of populations may be obscured if the scale of our observations is not appropriate to the underlying biological system. Furthermore, restoration activities may have unexpected results if they do not consider patterns of connectivity and local adaptation (Allendorf and Waples 1996). Consequently, understanding the structure and boundaries of populations is necessary to implement effective management strategies.

Spatial heterogeneity and connectivity among suitable habitats can substantially influence both the structure and the boundaries of a population. While outright barriers to dispersal bound populations, biotic and abiotic factors may restrict connectivity between

groups of individuals (Fagan 2002, Apps and McLellan 2006), leading to patterns of population isolation by ecological features of the landscape rather than purely by geographic distance (e.g. isolation by resistance, McRae 2006). In terrestrial environments, organisms can usually disperse in many directions across the landscape, and most landscape features change slowly through time. In contrast, streams are highly dynamic linear environments with limited connectivity between adjacent watersheds (Fagan 2002, Poole 2002, Cote et al. 2009). Under these conditions, heterogeneity in stream networks is expected to contribute strongly to population structure (Wiens 2002).

Despite the differences between streams and terrestrial environments, individuals that occupy spatially contiguous habitat within a stream network are typically assumed to represent a single population. This assumption is important, pervasive, and generally untested. Conceptually, there are several alternative scenarios to a randomly mating (e.g., panmictic) population that need to be considered. A single population may exhibit genetic isolation by distance (Carlsson and Nilsson 2000, Kanno et al. 2011) where gene flow is spatially restricted. In contrast, multiple discrete populations may be present (Carlsson et al. 1999). Finally, fish within a stream network may represent admixtures of genotypes originating from different local subpopulations within a regional metapopulation (Gotelli and Taylor 1999). Such alternative population structures may be widespread yet overlooked because of the linear nature of stream networks. In many cases, they can only be detected with modern molecular techniques.

These alternative population structures within a stream network require different approaches to management. For example, groups of populations may exhibit a portfolio effect, potentially obscuring the declines of individual populations (Schindler et al. 2010).

Even within a population, different life history strategies may have different geographic boundaries. For migratory fishes, this may result in aggregations of migratory individuals from different populations during non-reproductive periods, as has been observed in numerous salmonids (Keefer et al. 2006, Nyce et al. 2013). Furthermore, if local adaptation has occurred, populations could differ in key measurable traits with impacts to our understanding of population dynamics. These scenarios highlight the importance of characterizing the structure of populations so that we may more effectively understand a species and its surrounding landscape.

Brook Trout are well-suited to studies of population structure. The species is broadly distributed and well-studied yet exhibits considerable life history variation (Power 1980, Kennedy et al. 2003, Theriault and Dodson 2003) and genetic diversity (King et al. 2012, Aunins et al. 2014). Widespread declines have attracted attention, and millions of dollars are spent annually on the conservation and restoration of Brook Trout. These management activities have frequently been met with unexpected results (e.g. Richards 2007), potentially as the result of misunderstanding population structure and function.

Our ultimate goal was to explore the population structure of stream resident Brook Trout in a highly connected stream network in hopes of better understanding its function with respect to management. In this paper we test the hypothesis that Brook Trout in a connected stream network represent a single, panmictic population against the alternatives involving population differentiation on small spatial scales. Where differences existed, we further explored relationships in key measurable traits.

Methods

The Savage River watershed of western Maryland contains >100 km of interconnected Brook Trout habitats (Heft 2006) and has been identified as a regionally important population stronghold. Within this larger drainage, we focused on the Big Run watershed, which contains about 24 km of perennial streams supporting Brook Trout. The Big Run watershed contains one major tributary named Monroe Run, which forms a Y-shaped network (Figure 15). The study streams support a typical Appalachian Plateau stream community dominated by Brook Trout, Blue Ridge Sculpin (*Cottus caeruleomentum*), and Eastern Blacknose Dace (*Rhinichthys atratulus*). We concentrated our efforts on 4.5 km of stream closest to the downstream terminus with the upper Savage River.

Brook Trout were collected biannually in the summer (June-July) and autumn (September) using backpack electrofishing across the entire study area. Following capture, all individuals were chemically anesthetized (80 mg/L tricaine methanesulfonate buffered with 0.2 mM $NaHCO_3$, pH = 7), measured, and scanned for a passive integrated transponder (PIT) tag. During summer sampling, any previously un-captured individuals were implanted with 12 mm PIT tags. Adipose fin biopsies were obtained from each individual and preserved in 95% ethanol for genetics analysis.

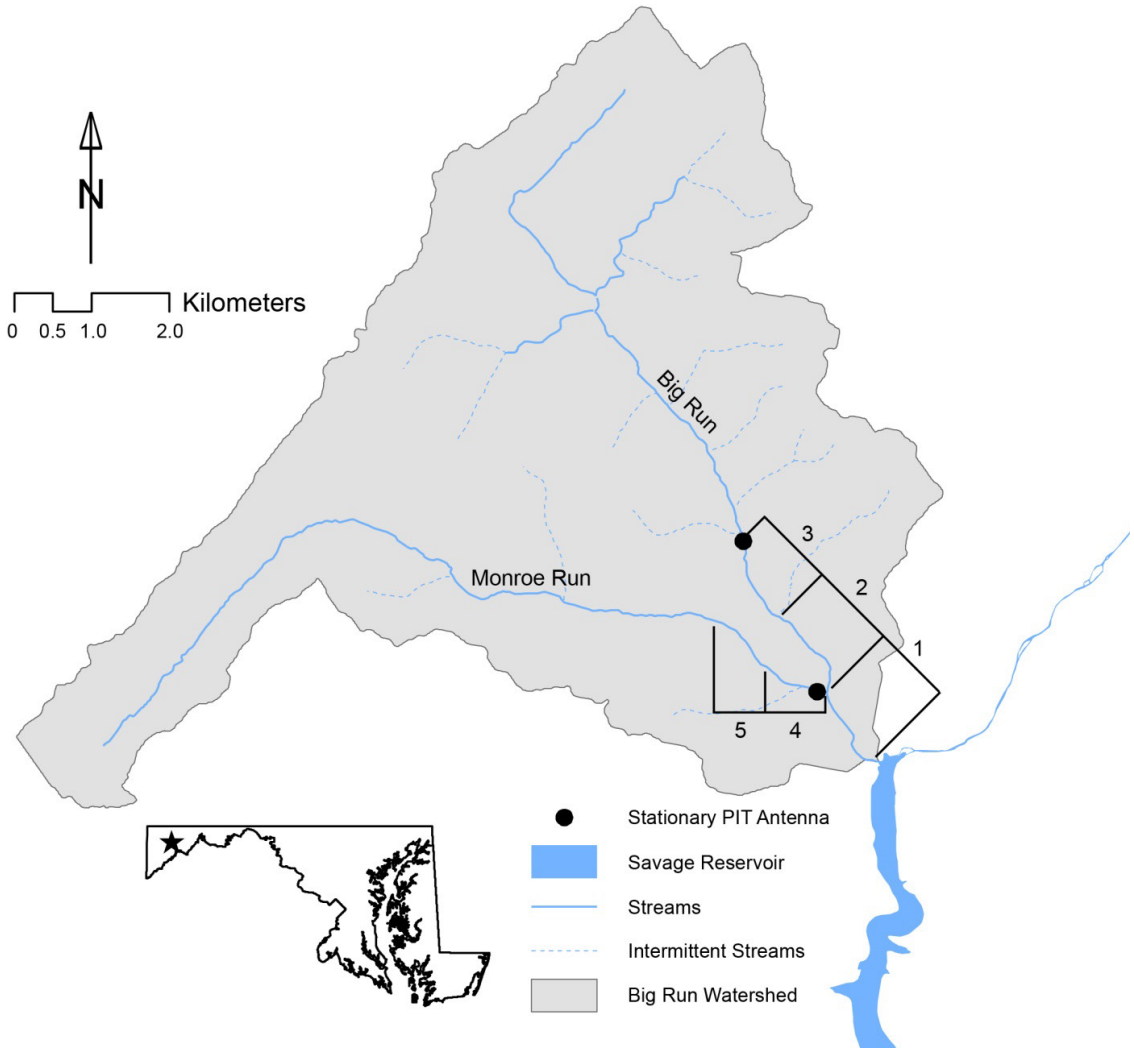

Figure 15. Our study focused on five contiguous sections of the Big Run watershed in western Maryland. Stationary passive integrated transponder (PIT) antennas were operated at two sites within the study area.

Population Genetics

We selected 250 Brook Trout from across the study area for genetic analysis. Individuals were chosen using a stratified random sampling design where about 50 individuals were selected from each of five stream reaches, representing a mixture of stationary and mobile individuals. An individual was considered to be mobile if it moved >0.1 km between physical recapture events.

Brook Trout were genotyped at 13 microsatellite loci using the methods described by King et al. (2012). One individual failed to yield a genotype at multiple microsatellite markers, and was omitted from the remainder of the investigation. Among the remaining individuals, we were able to obtain genotypes at 99.4% of loci (3219/3237 genotypes). Prior to any subsequent analysis, we used COLONY 2.0.5.0 (Jones and Wang 2010) to identify individuals with full-sibs represented in the sample. Where they occurred, full-sibs were randomly removed from the dataset (n = 24) until only a single family representative remained in each section. Removal of excess full sibs from the data set reduced the number of multilocus genotypes available within a section by 7.5-11.5%. Four hundred sixty-nine half sibling pairs were identified among the genotyped individuals, but were left in the data set for analysis.

We used GENALEX, version 6.501 (Peakall and Smouse 2006, Peakall and Smouse 2012), to determine if Brook Trout collected within a stream reach appeared to conform to Hardy-Weinberg equilibrium, based on a Bonferroni-adjusted critical P-value (0.0008). GENALEX was also used to calculate pairwise F_{ST} and G''_{ST} (Meirmans and Hedrick 2011) values among collections in different stream reaches in an analysis of molecular variance (AMOVA) framework (Weir and Cockerham 1984). Next, we used GENEPOP, version 4.2.1, to evaluate if there was linkage disequilibrium between loci within our stream reaches.

We used STRUCTURE 2.3.4 (Pritchard et al. 2000, Falush et al. 2003) to determine the most likely number of populations (K) present within the study area and examined the results using STRUCTURE HARVESTER (Earl and vonHoldt 2012). The number of clusters tested ranged from K=1 through K=8 with 20 iterations performed per

cluster. Each model run used correlated allele frequencies and considered the section of capture as a prior. We used CLUMPP 1.1.2 (Jakobsson and Rosenberg 2007) to align repeated model runs and generated ancestry plots using DISTRUCT 1.1 (Rosenberg 2004).

To quantify long-term rates of genetic exchange among groups, we implemented a Bayesian coalescent model in MIGRATE and estimated θ and M, where θ represents the mutation-scaled effective population size and M represents the mutation-scaled immigration rate (Beerli 2006, Beerli and Felsenstein 2001). Migration rates were allowed to be asymmetric and to vary between groups. A preliminary examination of allele frequencies found little support for the stepwise mutation model, so we used the Brownian motion option within MIGRATE and assumed a constant mutation rate across all loci. Prior distributions for θ and M were uniform between minimum and maximum values, set as 0-20 and 0-1000 respectively. The model ran with an initial burn-in of 10,000 trees, followed by data collection for 10,000 MCMC sweeps every 100 steps. Static heating was used (four chains) and the chains were allowed to swap. Median values from posterior distributions were used for parameter estimates. To calculate long-term genetic exchange rates in units of effective migrants from group j to group i, we used the relationship described by Beerli (1998; Equation 2).

$$Immigrants\ per\ generation = \frac{\theta_i M_{j \to i}}{4} \qquad \text{(Equation 2)}$$

Life History Differentiation

Based on recaptures of individually marked fish, we characterized the physical movement, growth, and survival patterns of Brook Trout within the study area:

Movement - We described movement patterns within our study area using mark-recapture observations and stationary PIT antennas. Stationary PIT antennas were operated at two sites within the study area to detect fish movements that would have gone undetected using only a mark-recapture study design (Figure 15; Kanno et al. 2014). One antenna array was located on Monroe Run (within section 4) approximately 50 m upstream of its confluence with Big Run. A second antenna array was located at the upstream extent of our study area on Upper Big Run (within section 3). At each location, two pass-by antennas constructed of high-density polyethylene were anchored to the stream bottom and operated between June 2011 and September 2013 (Kazyak and Zydlewski 2012). Although battery failures resulted in occasional periods where antennas were not operational, the data derived from these arrays should be a representative sampling of individuals who visited the antenna sites.

Growth - We calculated annual growth increments for individuals that were recaptured on consecutive summer electrofishing samples (Equation 3). We used analysis of covariance (ANCOVA) to compare observed growth rates between groups of brook trout while accounting for the effect of individual size for each of the three years of the study. Analysis of covariance models initially considered a size by group interaction term. Where the interaction term was not significant, the models were refit with only the main effects.

$$Annual\ growth = \frac{L_1 - L_0}{t_1 - t_0} * 365 \qquad \text{(Equation 3)}$$

Survival - Based on four years of electrofishing surveys, we generated individual encounter histories for 2,973 fish representative of the two streams (1,919 from Big Run and 1,054 from Monroe Run). Forty individuals were captured in both Monroe Run and Big Run during summer sampling events, and were omitted from survival analysis because their genetic association was uncertain. We used multistate Cormack-Jolly-Seber models implemented in program MARK (White and Burnham 1999) to estimate stage-specific survival rates for Brook Trout. Stages were defined based on total length: young-of-the-year (YOY; <100 mm based on field observations) and adult (A; ≥100 mm). During each annual time step, an individual could survive or die, and all surviving YOY transitioned (Ψ) into adults. Survival rates (*S*) were allowed to vary by year and by stage. Catchability (*p*) was assumed to be constant for each of the summer samples, but modeled independently for fall 2013, as this sample was based on a single pass of electrofishing whereas the other samples used three consecutive passes. We assumed all tags were retained and successfully read during recapture events.

Results

Population Genetics

We successfully obtained multilocus genotypes from 249 Brook Trout. Samples from within each of the five sections largely conformed to Hardy-Weinberg equilibrium (Table 5). However, in section 3, two loci showed a departure from Hardy-Weinberg equilibrium ($P < 0.0038$). Significant linkage disequilibrium was detected in one pairwise comparison of loci in section one and in two instances in section 5 (Table 5; $P < 0.0001$).

Table 5. Sample size before (N_{Total}) and after (N_{Analyzed}) redundant full-sibs were removed from the study, mean allelic richness (A), observed (H_o) and expected heterozygosity (H_e), number of private alleles, proportion of loci conforming to Hardy-Weinberg equilibrium (HWE), and the proportion of loci pairs in significant linkage disequilibrium (LD).

Section	N_{Total}	$N_{Analyzed}$	A	H_o	H_e	Private alleles	HWE	LD
1. Lower Big Run	40	36	7.692	0.701	0.693	1	13/13	1/78
2. Upper Big Run	52	46	7.462	0.695	0.697	4	13/13	0/78
3. Upper Big Run	53	49	7.769	0.693	0.702	1	11/13	0/78
4. Monroe Run	53	47	7.154	0.670	0.659	1	13/13	0/78
5. Monroe Run	51	47	7.154	0.705	0.685	1	13/13	078

Bayesian genetic clustering showed support for two genetic groups that inhabit the study area, based on comparisons of Delta *K* and model likelihoods for models considering K =1 through K =8 (Program STRUCTURE; Figure 16). Inferred membership of individuals in the two genetic clusters clearly reflected the geographic layout of the stream network, with Monroe Run fish consistently assigned to one group while Big Run fish upstream of the confluence with Monroe Run were consistently assigned to another (Figure 17). Genetic differentiation was small, but significant between streams (F_{ST} = 0.011-0.013 and G''_{ST} 0.029-0.047; Tables 6-7), and no differentiation was detected within streams. Overall, 1.22% of the observed molecular variance could be attributed to differences between upper tributaries (AMOVA). Downstream of the confluence of Monroe and Big Run, individuals were predominantly of Big Run origin, and only five fish had STRUCTURE membership in the Monroe Run cluster. A single individual in Upper Big Run (section 2) was not definitely assigned, but still received marginal support for assignment with the other fish from Upper Big Run (Q = 0.505).

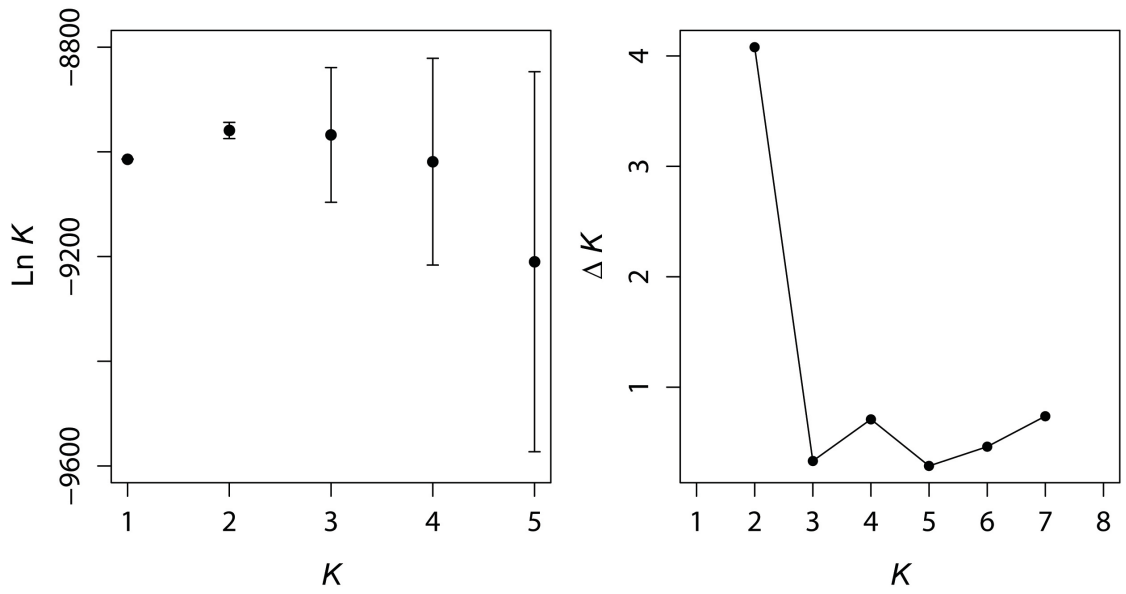

Figure 16. Mean log likelihood (±1 SD) calculated over 20 model runs at each K and ΔK, an *ad hoc* statistic proposed by Evanno et al. (2005) to estimate the correct number of clusters. Likelihoods for K 6 through 8 were omitted for graphical purposes, but were less than the values shown for K 1 through 5.

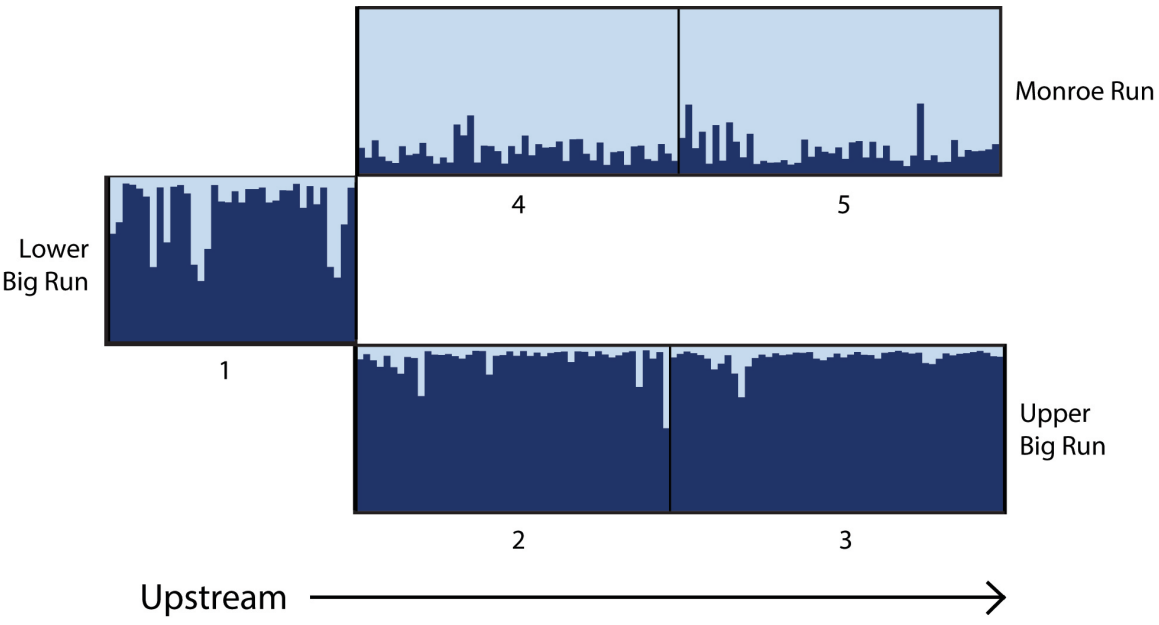

Figure 17. Individual assignment probabilities for individuals in five stream sections based on two clusters ($K = 2$) for Brook Trout in the Big Run watershed. Stream sections were considered as prior information and correlated allele frequencies were used.

Table 6. Pairwise F_{ST} values among Brook Trout collected in five study sections in the Big Run watershed. F_{ST} values are shown below the diagonal and *P*-values are shown above. Values that were statistically significant (based on a Bonferroni-adjusted critical *P*-value of 0.005) are shown in bold.

Section number and location	Section number				
	1	2	3	4	5
1. Lower Big Run	-	0.423	0.881	**0.005**	**0.004**
2. Upper Big Run	0.006	-	0.386	**0.002**	**0.001**
3. Upper Big Run	0.005	0.006	-	**0.001**	**0.001**
4. Monroe Run	0.011	0.012	0.013	-	0.293
5. Monroe Run	0.011	0.012	0.011	0.006	-

Table 7. Genetic divergence among Brook Trout collected in five study sections in the Big Run watershed. Pairwise G''_{ST} values are shown below the diagonal and *P*-values are shown above. Values that were statistically significant (based on a Bonferroni-adjusted critical *P*-value of 0.005) are shown in bold.

Section number and location	Section number				
	1	2	3	4	5
1. Lower Big Run	-	0.414	0.886	**0.003**	**0.003**
2. Upper Big Run	0.001	-	0.376	**0.000**	**0.000**
3. Upper Big Run	0.000	0.002	-	**0.000**	**0.000**
4. Monroe Run	0.031	0.041	0.049	-	0.316
5. Monroe Run	0.031	0.041	0.037	0.003	-

The long-term rate of genetic exchange between the two groups was low (39 effective immigrants per generation from Monroe Run into Big Run and nine effective immigrants per generation from Big Run into Monroe Run; Table 8). Assuming a three year generation time (see Chapter 5), annual immigration is 13 effective migrants into Big Run and three effective migrants into Monroe Run.

Table 8**.** Estimates of mutation-scaled effective population size (θ), mutation-scaled immigration rate (*M*), and number of effective immigrants for Brook Trout populations in Big Run and Monroe Run.

Population	θ_i	$M_{j->i}$	Effective immigrants/generation	Effective immigrants/year*
Big Run (Sections 2-3)	5.55	28.33	39.34	13.11
Monroe Run (Sections 4-5)	1.58	23.67	9.35	3.12

**Based on an average generation time of three years.*

Life History Differentiation

Based on evidence for two distinct groups occupying adjacent streams, subsequent life history trait analyses were applied separately to Big Run (sections 1-3) and Monroe Run (sections 4-5) Brook Trout.

Movement - Brook Trout in the two streams were clearly distinguished by movement patterns, and exchange of individuals between the two streams was rare based on the electrofishing survey data. Brook Trout initially tagged in Monroe Run (n = 1,058) were never captured during electrofishing surveys in Big Run above the confluence (Table 9). Furthermore, trout tagged in upper Big Run (n = 1,443) were very rarely detected in Monroe Run (n = 4), and three of the four detections were within 50 m of the confluence. Among fish tagged in either stream, 99.2% of relocations occurred within the same stream. When a fish left the stream of initial capture, it was found below the confluence in 12/16 instances (Table 9). Furthermore, based on recaptures during consecutive fall sampling events, very few fish moved between the tributaries from year to year (Table 10). To summarize, exchange of Brook Trout between the two streams was rare based on the electrofishing survey data.

Table 9. Distribution of relocations of Brook Trout initially tagged during the summer in five contiguous stream reaches based on physical recaptures and stationary passive integrated transponder antennas. The number of fish tagged in each reach is denoted by *n*.

			Number of physical recaptures					Number of individuals detected	
Section	Description	*n*	1	2	3	4	5	Big Run Array	Monroe Run Array
1	Lower Big Run	513	277	46	6	14	3	30	42
2	Upper Big Run	686	4	514	64	3	0	75	6
3		757	0	35	486	1	0	182	2
4	Monroe Run	447	7	0	0	390	69	3	60
5		611	1	0	0	8	523	1	16

Table 10. Estimates of migration rates between Big Run and Monroe Run based on mark-recapture data for individuals collected in consecutive fall sampling events and summer population estimates.

Population	N	Emigration (%)	Immigrants/year
Big Run (Sections 2-3)	687	0.70%	9.13
Monroe Run (Sections 4-5)	377.25	2.42%	4.77

Movement patterns recorded at the two stationary antennas were similar to those inferred from our mark-recapture observations. The stationary PIT antenna arrays recorded >160,000 tag detections representing 413 of 3014 (13.7%) of the tagged individuals. Most of the detections were due to a few individuals that were repeatedly detected. The antennas at the downstream terminus of Monroe Run recorded 126 unique fish that comprised primarily individuals whom were tagged in Monroe Run (n = 76), and to a lesser extent, individuals that were tagged in Lower Big Run (n = 42). Fish that were initially tagged in Upper Big Run (n = 8) were very seldom recorded on the Monroe Run antennas. The antennas at the upper extent of the study area on Upper Big Run recorded 291 unique Brook Trout. These individuals were mostly tagged on Upper Big Run (n =

257) and Lower Big Run (n = 30), with only a few trout originating in Monroe Run (n = 4). The majority of tag detections at both arrays represented individuals that had been tagged in sections adjacent to the stationary antennas (68.1%). Overall, the observed movement patterns suggest a large degree of isolation between the two streams. Where exchange occurs, it is generally individuals transitioning between Lower Big Run and one of the tributaries, rather than between the two tributaries.

Growth - Annual growth increments ranged from 0 to 119.9 mm, and decreased linearly with respect to body size in each year ($P < 0.05$). Despite substantial interannual variation, we found significant differences in growth rates (ANCOVA*;* $P < 0.05$) between individuals in Big Run and Monroe Run for each of the three years of record. Brook Trout in Big Run consistently grew faster (overall 7.8 mm/y at a given length; $P < 0.05$) than in Monroe Run (Figure 18).

Survival - Estimated annual survival rates ranged from 31.0-45.0% for adults, with no significant difference in year-specific survival rates between the streams (Figure 19). Similarly, young-of-the-year survival rates ranged from 27.2-42.0% and were similar among the streams in 2011 and 2012 (Figure 19). Unfortunately, low recruitment in 2010 prohibited meaningful estimates of young-of-the-year survival rates for both streams.

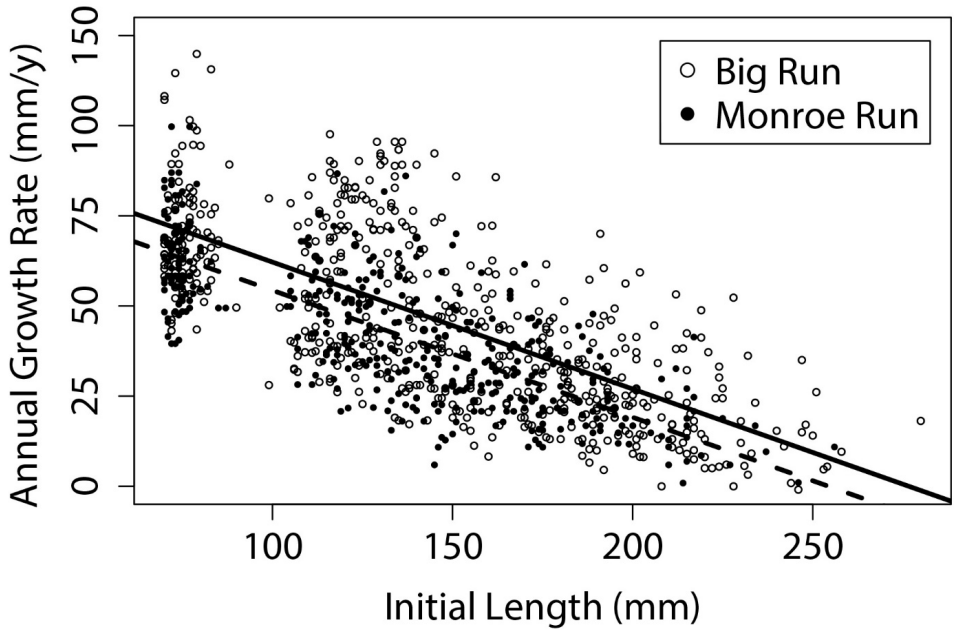

Figure 18. Observed annual growth rates for Brook Trout in Big Run and Monroe Run, 2010-2013. The lines represent mean annual growth for individuals of a given length from Big Run (solid) and Monroe Run (dashed).

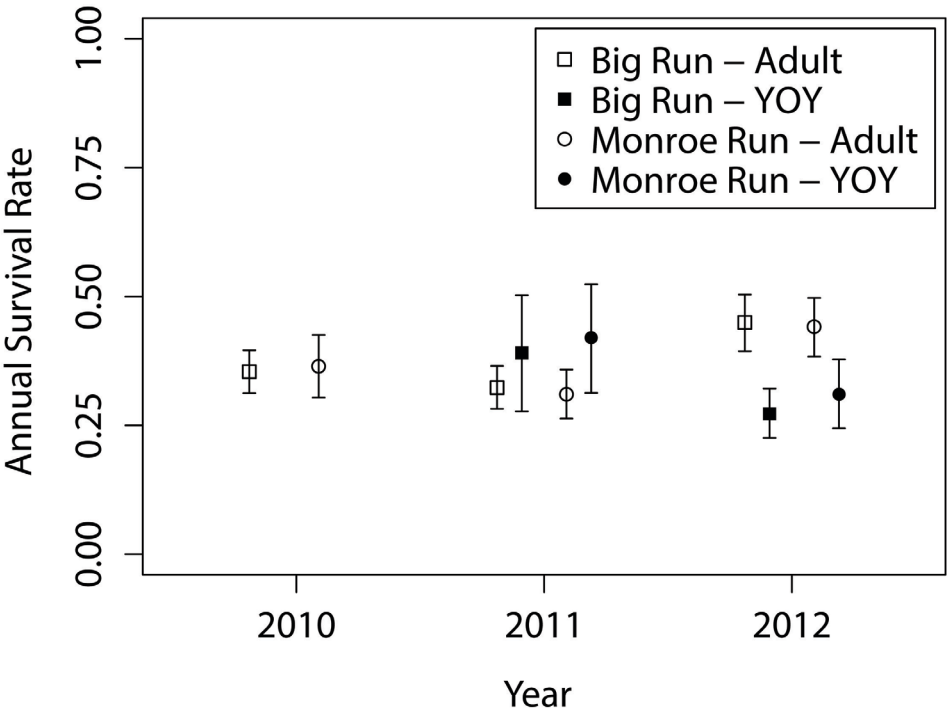

Figure 19. A comparison of parameter estimates derived from multistate Cormack-Jolly-Seber models fit to fish in Big Run and Monroe Run using program MARK. There were insufficient numbers of young-of-the-year (YOY) fish collected in 2010 to generate meaningful estimates of survival rates.

Discussion

The Brook Trout within the Big Run watershed appear to form two genetically distinct groups inhabiting adjacent and connected streams. The movement patterns exhibited by these groups appear to limit the potential for genetic exchange between streams, reinforcing the observed genetic structure. However, the estimated long-term rates of genetic exchange between the two streams shows some degree of connectivity. Thus, exchange occurs between streams, but introgression rates are limited to a few immigrants per generation.

We believe the Brook Trout in the study system represent a cryptic metapopulation: a spatially continuous distribution of organisms exhibiting metapopulation-like behaviors. Although Levins (1969, 1970) focused on extinction-recolonization dynamics in his model of metapopulations, later adaptations relaxed the definition to include groups of local populations that function largely autonomously but have a nontrivial exchange of individuals (Hastings and Harrison 1994, Kritzer and Sale 2004). Our study presents evidence of the latter scenario in lotic Brook Trout on a small spatial scale and in the absence of barriers to migration. Despite decades of population monitoring, the metapopulation-like structure of Brook Trout within the Big Run watershed remained unnoticed.

Increasingly, complex population structure has been revealed in many groups of organisms that were previously thought to represent single populations (Brattström et al. 2010, Hess et al. 2011, Hindrikson et al. 2013). In many cases, these organisms have a high dispersal capability or live in continuous habitat, leading to an expectation of panmixia (Palumbi 1994). In streams, where dispersal is often limited and habitats are highly heterogeneous, we should expect population differentiation to be even more

prevalent. Although some migration barriers such as waterfalls or culverts are readily identified, other factors that influence dispersal and genetic exchange may be less obvious. Water chemistry and temperature, both of which vary continuously through time, may restrict or prohibit genetic exchange within stream networks (Aunins et al. 2014). If such isolation by ecological resistance is commonplace, then cryptic metapopulations may be widespread, yet have largely remained undetected.

The existence of cryptic metapopulations has implications for the ecology of stream fishes. These population structures may serve to enhance resiliency by stabilizing abundance through time, as in the context of a portfolio effect (Schindler et al. 2010). Undetected structuring within populations may also help explain the observed life history variation in many species. The presence of cryptic metapopulations may also increase standing genetic diversity that can facilitate future adaptation (Barrett and Schluter 2008). Thus, cryptic metapopulations may influence both the contemporary population dynamics and future adaptive potential of stream fishes.

Within a stream network, patterns of local selection and limited connectivity may lead to population differentiation. Among salmonids, local adaptation is relatively common and sometimes evident on small spatial scales (Fraser et al. 2011). Local adaptation may be manifest in many traits, including morphology (Quinn et al. 2001), thermal response (Meier et al. 2014), movement patterns (Letcher et al. 2007), and vital rates (Koskinen et al. 2002, Letcher et al. 2007). Interestingly, although Big Run and Monroe Run appeared to be offer similar habitats (Table 11), we found evidence for consistent differences in growth rates. In Brook Trout, temperature and food availability are the two primary drivers of growth. Over the course of two years, the two streams

Table 11. Comparison of key habitat metrics among Lower Big Run, Upper Big Run, and Monroe Run. Maximum depth, residual pool depth, and pool coverage are presented as an average value for a 50 m reach ± 1SD. Temperature data were derived from loggers deployed at fixed sites within the study area from summer 2012 through spring 2014 and are reported as a mean ± 1SD. Water chemistry data was collected by the Maryland Biological Stream Survey in 1996 (Big Run; GA-A-090-310-96) and 2000 (Monroe Run; SAVA-101-C-2000).

Habitat metric	Lower Big Run (1)	Upper Big Run (2-3)	Monroe Run (4-5)
Total length (km)	0.95	2	1.5
Wetted width (m)	6.0 ± 2.0	4.4 ± 1.5	3.6 ± 1.2
Maximum depth (cm)	60.2 ± 36.0	39.1 ± 18.4	34.9 ± 12.3
Residual pool depth (cm)	42.9 ± 32.3	27.9 ± 18.3	23.5 ± 11.8
Pool (%)	18.9 ± 14.9	12.8 ± 11.3	11.7 ± 11.8
Mean seasonal temperature (ºC)			
Winter	1.9 ± 1.9	2.2 ± 1.8	1.8 ± 1.8
Spring	9.9 ± 4.1	9.6 ± 3.8	9.9 ± 4.1
Summer	17.0 ± 1.9	16.5 ± 2.0	16.9 ± 2.1
Fall	7.2 ± 4.3	6.9 ± 4.1	6.7 ± 4.3
Maximum temperature (ºC)			
2012	21.6	21.7	21.9
2013	20.5	19.2	20.4
Water chemistry			
pH	-	7.06	7.15
Conductivity (µS/cm)	-	50	70
Nitrate (ppm)	-	0.5	0.3
Sulfate (ppm)	-	11.8	12.3
Dissolved organic carbon (ppm)	-	0.9	1.1

experienced nearly identical thermal regimes, and we do not expect temperature was a significant factor in the observed growth dichotomy. We are unfortunately unable to explicitly compare productivity between the two streams, although the habitats are very similar. Regardless of the drivers of growth, the observed discrepancy in growth rates may result in differences in age-specific reproductive output. Conversely, stage-specific survival rates were very similar for the two streams in each year. Thus, we lack compelling evidence to conclude that large differences in selection pressures are driving differentiation.

Tributary confluences are often pronounced zones of ecological change (Poole 2002, Benda et al. 2004, Kiffney et al. 2006), and these are sometimes associated with genetic boundaries (Carlsson et al. 1999, Neville et al. 2006) or clines (Kanno et al. 2011). The movement patterns we observed may promote reproductive isolation and help maintain the observed population structure. Despite the absence of barriers to movement within the study area and the potential mobility of Brook Trout, we observed limited movement overall. Among the fish that were mobile, individuals very seldom moved between the two tributary reaches. We lack evidence to know if individuals that moved into Lower Big Run returned to tributaries to spawn, but we speculate that little genetic exchange occurs among the two groups of Brook Trout where they co-occur in Lower Big Run. Based on fall spawning surveys, reproductive effort is minimal in Lower Big Run relative to its tributaries. This lack of spawning activity may enhance the genetic separation between tributaries, since this is the only area where we found much spatial overlap between the genetic groups. Positive assortative mating may further isolate the genetic groups. In a stream where three strains of Brook Trout were stocked in sympatry,

Richards et al. (2007) found the majority of reproduction arose from same-strain matings. Thus, mechanisms for isolation exist among some Brook Trout strains and this might help to explain the large degree of genetic divergence found throughout its native range (Aunins et al. 2014, King unpublished data). An alternative mechanism for isolation may involve natal homing that limits straying among tributaries during the spawning season, thus moderating the effects of seasonal dispersal. We speculate that Brook Trout may use olfactory cues to guide movement patterns and reproductive behavior, ultimately contributing to reproductive isolation. This behavior is well-documented in Pacific salmon (Wisby and Hasler 1954, Scholz et al. 1976, Keefer et al. 2006), and may be broadly conserved across stream fishes (Hasler and Wisby 1951, Armstrong and Herbert 1997).

Although new analyses may show cryptic metapopulations to be widespread in streams, it may be unrealistic to manage such taxa. The scale and scope of the effort required to identify and individually manage these population components is simply not feasible in most cases, particularly if they are not spatially segregated. However, it is important to recognize and conserve the underlying population structure, as the components may represent local adaptation and reservoirs of genetic diversity. When fishes are extirpated from a stream reach within a larger watershed, genetically distinct populations may be wiped out, even if other conspecifics later recolonize the habitat. Additionally, restoration and reintroduction activities may be more successful if the population structure is understood, especially when dealing with species known for high amounts of genetic divergence. For example, a dwindling population may be more difficult to restore by improved connectivity or manual transfers (Hilderbrand 2003) if

fishes exhibit population divergence on a small geographic scale because the underlying mechanism for divergence may inhibit interbreeding. Further, efforts to improve connectivity may be overstated if fish populations have naturally evolved to persist in small patches, even where larger areas of interconnected habitat are available.

Cryptic metapopulations may play an important role in the ecology of stream fishes, but are easily overlooked where organisms are continuously distributed. Future work should seek to determine the prevalence of cryptic metapopulations and seek to understand their roles in stream ecology. An improved understanding of this hidden level of population organization may help to improve management and restoration outcomes.

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Chapter 5: Brook Trout population dynamics under current and alternative conditions

Abstract

Environmental stochasticity is a ubiquitous force driving the population dynamics of fish and wildlife populations. The vital rates of populations commonly vary in response to environmental conditions, which are predicted to rapidly change during the next century. Although the causes of environmental change are outside of the traditional scope of fisheries management, managers may be able to mitigate some of the impacts of climate change using established regulatory approaches. Given the pressing need for action and difficulty of evaluating management strategies *in situ*, simulation models may offer insight into the range of expected outcomes if environmental conditions or management strategies where to change. We used a large-scale tagging study to parameterize stochastic projection models and explore how different management strategies might influence population resilience and persistence under several environmental regimes. We found population dynamics were driven by pulse-driven recruitment that was only weakly related to spawner abundance. Simulated changes in adult survival, representative of a range of management scenarios, had a considerable impact on population resilience. Conversely, changes in the growth rates of Brook Trout resulted in small changes to population resilience. None of the simulations resulted in extinction after 50 years. Enhanced adult survival resulted in a greater abundance of large fish. Collectively, these results suggest regulatory approaches may offer some utility in

promoting population resilience while enhancing the quality of the fishery, but are likely insufficient to fully offset the impacts of predicted environmental changes.

Introduction

Environmental variation is a ubiquitous force driving the dynamics of fish and wildlife populations. The vital rates of populations commonly vary in response to environmental conditions. Growth rates of ectotherms can vary substantially with even small changes in temperature (Brett et al. 1969, Deeming and Ferguson 1989, Brylawski and Miller 2006). Periods of unusual temperatures or precipitation can trigger major mortality events and recruitment failures (Hakala and Hartman 2004, Elliot et al. 1997, Foley et al. 2008). Variability in circulation patterns can have strong effects on the transport and abundance of fishes (Norcross and Shaw 1984, Caputi et al. 1996), marine invertebrates (Caputi et al. 1996), and insects (Drake and Farrow 1983, Joyce 1983). Even in ostensibly stable environments, small environmental changes can have measurable impacts on the population dynamics of organisms (Anderson and Deacon 2001).

The relative importance of environmental stochasticity on population dynamics presumably varies with population characteristics and the predictability of the environment. Animals with short generation times, restricted habitats and mobility, and sensitivity to environmental conditions are expected to be especially influenced by variability in the environment. Stream dwelling fish are expected to be particularly sensitive to environmental variability, due to their limited dispersal potential and the highly dynamic nature of streams (Fagan 2002, Poole 2002).

Weather patterns are widely predicted to become increasingly harsh and variable in the context of global climate change. In the northeastern United States, mean annual temperatures have risen by 2ºC since 1970, and are forecast to rise several additional degrees over the next few decades (Karl et al. 2009). Furthermore, the frequency of extreme weather events is increasing - a trend that is expected to continue or worsen (US EPA 2001, IPCC 2001). These changes have already had a measurable impact on the ecology and abundance of many organisms, and are predicted to further increase the variability in their population dynamics (Walther et al. 2002). Thus, identifying effective management strategies becomes increasingly more difficult. Management challenges are even greater when dealing with exploited species and allowing for harvest while minimizing risk.

Brook Trout are an excellent model organism for studying the influence of environmental stochasticity in population dynamics. Although Brook Trout exhibit plastic life history strategies (Power 1980), they have experienced widespread declines (Hudy et al. 2008). Many of the remaining populations are found in headwater streams, where environmental variability can be considerable. Unfavorable environmental conditions can lead to multiple years of low recruitment. Brook Trout are especially sensitive to temperature and stream flow (McCormick et al. 1972, Stranko et al. 2008, Xu et al. 2010), both of which are expected to change under climate change projections (Milly et al. 2002, van Roosmalen et al. 2007, Isaak et al. 2012). However, despite these challenges, Brook Trout remain an iconic sportfish sought after by many anglers.

Although the many drivers of population declines are largely outside the scope of traditional resource management (i.e., climate warming, historic land conversion),

identifying intrinsic factors promoting population resilience may provide insights into actions that managers can institute. For example, fishing regulations may be implemented to promote survival and increase spawning stock biomass, thus speeding the recovery of a population from catastrophic events. If effective, such an approach offers an attractive opportunity for managers to mitigate some of the predicted impacts of climate change. Unfortunately, testing the effectiveness of these strategies is often infeasible due to the inherent costs, risks, and uncertainty involved with population scale manipulations of wild organisms. Furthermore, the immediacy of climate change impacts on fish and wildlife populations is becoming increasingly important as climate change adaptation strategies for coldwater fishes are needed now. Where experimental manipulations are not tractable, simulations studies can offer insight on the relative efficacy of management strategies in a dynamic world (Post et al. 2003).

Given what we already know and the uncertain future, how can we best leverage our limited abilities to promote population persistence and resilience given forecasts of the future? Our approach was to use a large-scale tagging study to parameterize stochastic population projection models to forecast alternative futures for Brook Trout. Our objectives were to explore how different management strategies might influence population attributes, resilience, and persistence under several potential environmental futures.

Methods

Alternative futures were forecasted for wild Brook Trout populations in the upper Savage River watershed of western Maryland (Figure 20). This watershed (approximately 300 km^2) represents a regionally important stronghold for Brook Trout, encompassing

multiple populations spread across >100 km of interconnected streams. Historically (1989-2006), anglers were allowed to harvest two Brook Trout per day in tributaries to the upper Savage River, with no terminal tackle restrictions. In response to population declines and perceived vulnerability, 'no-kill' regulations were implemented across the study area in 2007, which prohibit the use of live bait or the harvest of Brook Trout.

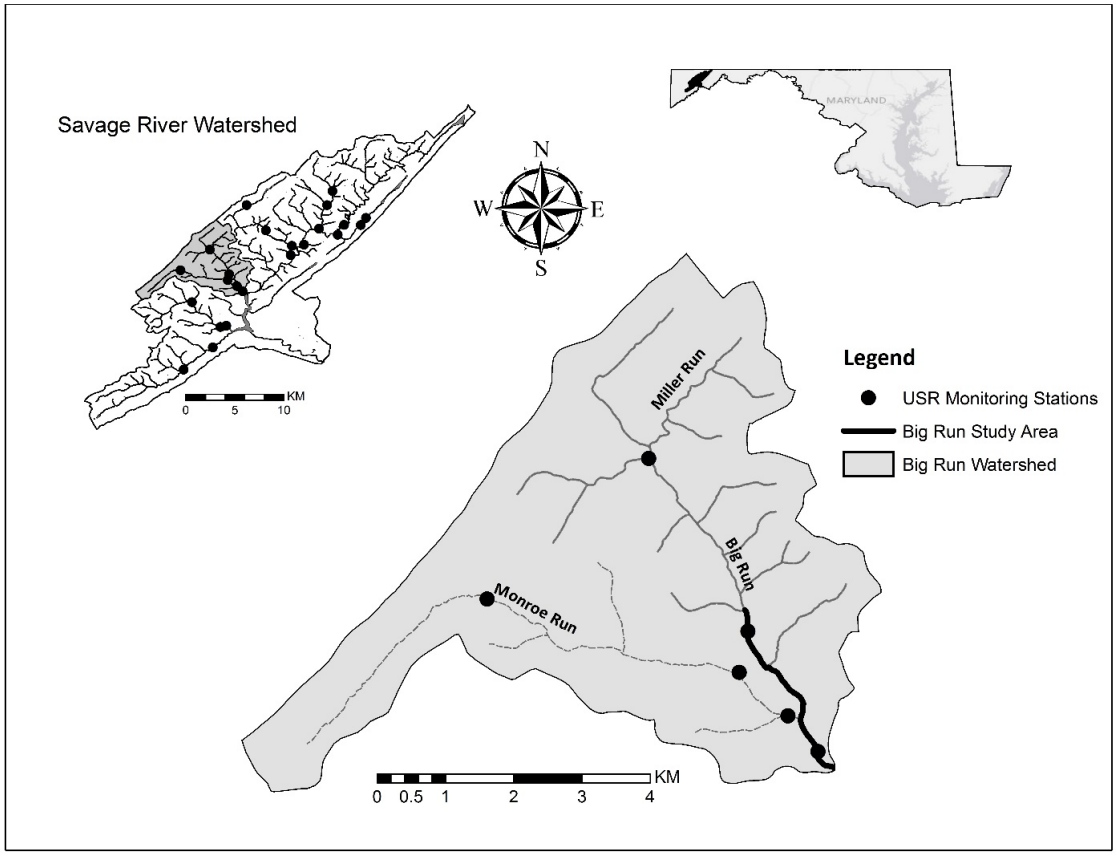

Figure 20. Our study focused on the population dynamics of Brook Trout in Big Run (shaded), a small forested watershed in western Maryland. We used mark-recapture data from the lower 3 km of Big Run (thick line) to parameterize growth and survival rates for simulation modeling. Recruitment patterns were estimated using long-term (2006-2014) monitoring data collected at 21 sites across the upper Savage River watershed (top left) by the Maryland Department of Natural Resources. Figure credit: Matthew Sell

We used two data sources to characterize the population dynamics of Brook Trout in Big Run, an upper Savage River tributary: (1) a long-term monitoring effort with census data from a network of sites across the upper Savage River watershed and (2) an intensive mark-recapture study focused on a single stream.

The long-term monitoring program was established in 2006 to offer insight into the status and trends of wild Brook Trout populations and their responses to the "no kill" regulations. Surveys were conducted at three stations on seven different tributaries in the upper Savage River watershed during the summer. At each site, multiple-pass removal surveys were conducted using backpack electrofishing gear on a 75 m reach. All Brook Trout were enumerated, individually measured (total length), and released.

The mark-recapture study was restricted to the Big Run watershed (39.5493ºN 79.1450ºW; Figure 20), a small, mostly forested catchment that drains into the upper Savage River. We conducted biannual electrofishing surveys of the entire study area (4.5 km) from 2010-2013. Brook Trout were anesthetized with tricaine methanesulfonate (80 $mg \cdot L^{-1}$; buffered with 0.2 mM $NaHCO_3$, pH = 7), measured (total length), and tagged with passive integrated transponder (PIT) tags. Following recovery, all fish were released within 50 m of their capture location. Observations from the mark-recapture study area are assumed to be representative of the entire watershed (15.5 km).

We used the field observations to parameterize stochastic stage-based matrix population models (Caswell 2001) representing current and alternative conditions. We divided Brook Trout into four stages based on total length: young-of-the-year (<100 mm; YOY), small (100-149 mm; S), medium (150-199 mm; M) and large (≥200 mm; L), selected subjectively based on the observed length-frequency distribution of Brook Trout

in Big Run and a reported relationship between length and fecundity (Letcher et al. 2007). For each scenario, we ran 1000 stochastic simulations and forecast the population trajectory for 50 years, using an adaptation of the popbio package (Stubben and Milligan 2007) in program R (R Development Core Team 2012). Each simulation was seeded with an initial population and forecast forward in one year increments. During each time step, individuals could survive or perish. Surviving individuals might remain in the same stage or grow to a larger one.

Transition matrices were generated based on two processes, growth and survival, which we assumed were independent. For each year, growth rates were empirically derived from observed growth trajectories. Annual survival rates were estimated using a multi-state Cormack-Jolly-Seber (CJS) model implemented in Program MARK (White and Burnham 1999). Two stages were defined: young-of-the-year (YOY; <100 mm) and adult (≥100 mm; A). The transition probability from YOY to A was fixed at 100%, based on our mark-recapture estimates of growth rates and an examination of length-frequency histograms. Survival was assumed to vary by year and stage, and estimated using maximum likelihood. To account for any bias due to tag loss, we adjusted our CJS estimates of survival (Pollock et al. 1990; Tables 12-13). The bias corrected estimates of survival were used for all subsequent analyses. For each year, the probability of an individual entering a given stage was calculated as the probability it would grow to that stage multiplied by probability it would survive to the next year, based on its stage at the start of the projection interval.

Table 12. Equations used in the parameterization and execution of Lefkovich population models.

Number	Equation	Description
1	$\frac{\hat{\phi}_{CJS}}{1-\hat{\theta}} = \hat{\phi}_{adj}$	Tag retention correction on survival estimates
2	$\begin{bmatrix} 0 & F_S & F_M & F_L \\ G_{S,YOY} & P_S & 0 & 0 \\ G_{M,YOY} & G_{M,S} & P_M & 0 \\ 0 & G_{L,S} & G_{L,M} & P_L \end{bmatrix} \begin{bmatrix} YOY_t \\ S_t \\ M_t \\ L_t \end{bmatrix} = \begin{bmatrix} F_t \\ S_{t+1} \\ M_{t+1} \\ L_{t+1} \end{bmatrix}$	Matrix projection model
3	$YOY_{t+1} = \alpha F_t e^{(\beta F_t + e^{\varepsilon})}$	Ricker stock-recruitment model

Table 13. Symbols representing model terms in this study.

Parameter	Description
Survival rates	
$\hat{\phi}_{CJS}$	Annual survival estimate derived from Cormack-Jolly-Seber model
$\hat{\phi}_{adj}$	Annual survival rate adjusted for tag retention
$\hat{\theta}$	Tag loss probability (0.017)
Stages	
YOY_t	Young of the year abundance at time t (≤100 mm)
S_t	Small abundance at time t (100-149 mm)
M_t	Medium abundance at time t (150-199 mm)
L_t	Large abundance at time t (≥200 mm)
Transitions	
P_i	Probability of surviving and remaining in stage i
$G_{j,i}$	Probability of surviving and growing from stage i to stage j
F_i	Fecundity for stage i
Ricker stock-recruitment model	
F_t	Total fecundity at time t
α	Density-independent term (0.0584)
β	Density-dependent term (-4.48×10^{-5})
ε	Recruitment error (log-scale standard deviation; 0.763)

We estimated egg production using a published length-fecundity relationship for Brook Trout collected in mountain streams of western Massachusetts (Letcher et al. 2007). This relationship was used to examine the relationship between egg production and recruitment across the 21 long-term monitoring sites (2006-2014), assuming a 1:1 sex ratio among the spawners. To account for low catchability of YOY (Borgstrom and Skaala 1993), we assumed YOY density was double the survey catch.

Recruitment in our projection models was estimated using a Ricker stock-recruitment model (Table 12-13) fit to the field observations of egg production and subsequent YOY recruitment. The model incorporated a lognormal error structure to reflect natural processes that contribute to recruitment variability.

We used 13 years of survey data from Big Run (1988-2014) to estimate carrying capacity of adults and YOY, assuming that the highest observed densities represented 90% of the carrying capacity of the system and the sex ratio was 1:1. Fish abundance was limited to these carrying capacities.

Our field-based estimates of life history parameters were used to parameterize a base model representative of recent conditions within Big Run (Figure 21). Using this model as a reference, we ran a series of simulations based on the premise that environmental change may change the relative frequency of fast and slow growth years. We considered four growth scenarios: (1) a reduced frequency of poor growth years, (2) the observed growth patterns continue, or (3-4) an increased frequency of poor growth years. (Table 14). For the purposes of our scenarios, the two years with the slowest growth rates were considered to be poor growth years.

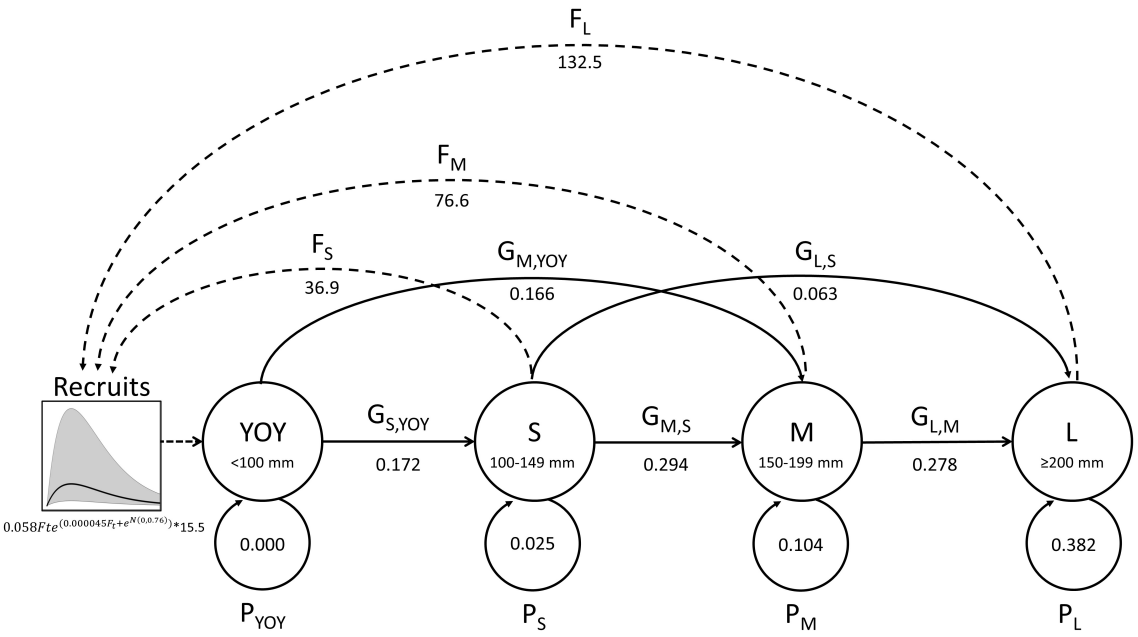

Figure 21. Conceptual representation of matrix projection model, with average values from the base model (B2) shown. Recruitment was estimated via a Ricker stock-recruitment model based on egg production and incorporated a lognormal error structure.

Table 1414. Growth scenarios considered for simulations of Brook Trout population dynamics.

Scenario	Description	Implementation
1	Enhanced growth	Reduced frequency (x1/2) of poor growth years
2	Status quo	No change
3	Reduced growth	Increased frequency (x2) of poor growth years
4	Greatly reduced growth	Increased frequency (x4) of poor growth years

To examine how changes in management might promote or inhibit population resilience, we considered four alternative scenarios with plausible yet differing patterns of adult mortality: (A) further angling restrictions (e.g. a moratorium) increase Brook Trout survival (+10%), (B) observed survival patterns continue, and (C-D) increased angling effort or changes in tackle restrictions result in reduce adult survival (-5% or -

10%; Table 15). Although we were primarily interested in how management can influence population resilience, these scenarios could also be interpreted to represent changes to adult survival due to environmental change.

Table 15. Survival scenarios considered for simulations of Brook Trout population dynamics.

Scenario	Description	Implementation
A	Enhanced survival	+10% to adult survival
B	Status quo	No change
C	Reduced survival	-5% to adult survival
D	Greatly reduced survival	-10% to adult survival

Population persistence was evaluated in each of the 16 scenarios by comparing the probability a simulated population would be extinct within 50 years, using the abundance of Brook Trout observed in 2014 as an initial population. We compared the relative resilience of populations by comparing their rate of recovery to a set threshold following a major disturbance event that reduced numbers to two females per 100 m of habitat (200 S, 60 M, and 50 L). The recovery threshold was reached when the population reached the median adult abundance observed in the base scenario (B2). Two-way analysis of variance (ANOVA) was used to formally contrast the recovery time for each of the scenarios. For the purposes of comparing scenarios, we also calculated the population growth rate (λ), generation time (Caswell 2001; Equation 5.73), and abundance by stage for each time step of the simulations. Finally, we examined the importance of recruitment patterns in population resilience and persistence by conducting an additional set of simulations under scenario B2, assuming a 30% reduction in recruitment relative to patterns observed in the monitoring data.

Results

Model Parameterization

We tagged 1955 Brook Trout in Big Run during electrofishing surveys from 2010-2013 (Table 16). Among these individuals, 27.6% were subsequently recaptured (776 total recapture events). Tag retention was estimated to be 98.3%.

Table 16. Number of fish tagged and recaptured during each electrofishing survey. Bold text denotes newly tagged individuals.

Tag Year	Cumulative recapture rate	2010	2011	2012	2013	Final
2010	25.5%	**770**	186	38	24	11
2011	33.2%		**575**	158	88	56
2012	25.1%			**610**	123	92

We observed considerable interannual variability in growth rates (Figure 22). During sampling intervals 2010-2011 and 2012-2013, growth rates were relatively slow. In contrast, we observed very rapid growth trajectories during 2011-2012. Across all years, we documented considerable variation among individuals with respect to growth. Additionally, many of the large fish (≥200 mm) exhibited very little annual growth (≤25 mm per year). For the purposes of simulation modeling, 2010-2011 and 2012-2013 were considered to be poor growth years.

Survival varied somewhat from year to year and among stages. Adult survival averaged 38.2% per year (range: 32.9-45.8%). Young-of-the-year survival averaged 33.8% (range: 27.8-39.8%). Unfortunately, a lack of available fish to tag precluded a survival estimate for YOY Brook Trout during 2010-2011. Consequently, the mean value from the other two years was used to populate the matrix of survival values for 2010-2011.

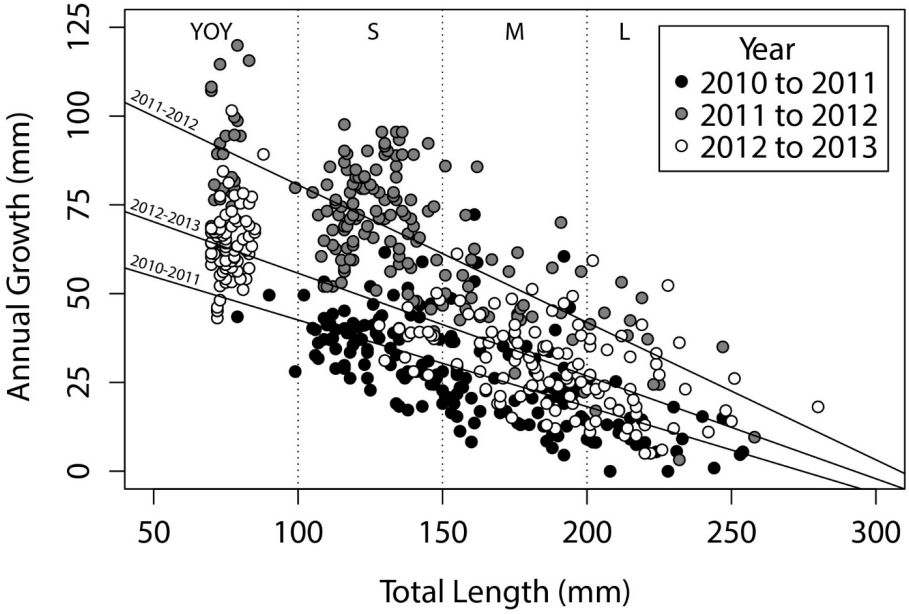

Figure 22. Annual growth rates of Brook Trout for three years of observation. Solid lines represent linear regression models fit to each of the years. Dotted lines delineate the stages used for simulation modeling.

The Maryland Department of Natural Resources captured 10,731 Brook Trout during nine years of monitoring at 21 sites on seven tributaries to the upper Savage River (Figures 20 and 23). Most fish collected were young-of-the-year (37.0%). The adults were mostly small (29.7%), with some medium (20.8%), and relatively few large fish (12.5%). Across the network of monitoring stations, the surveys documented four consecutive years of poor recruitment (2008-2011). Large numbers of young-of-the-year Brook Trout were observed in 2012 and 2013, followed by a marked rebound in adult abundance in recent years (2013-2014). On average, 267 female recruits were produced annually per km of stream, but years with poor recruitment were more common than those with excellent recruitment (Figure 24). The Ricker stock-recruitment model had a weak ($R^2 = 0.043$) but statistically significant ($P < 0.05$) fit to the field observations. Most of the variation in recruitment appeared to be independent of stock size and was well described by a lognormal error structure ($\sigma = 0.763$).

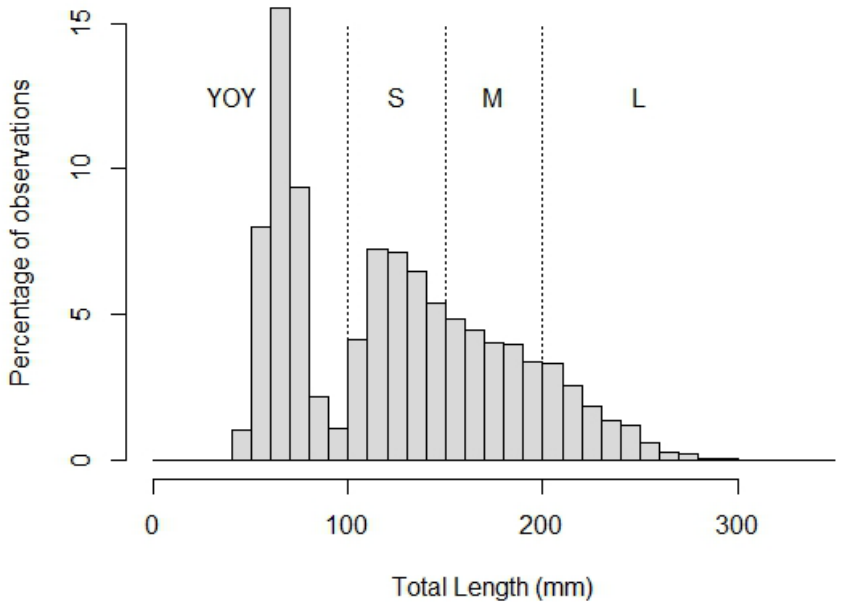

Figure 23. Size structure of Brook Trout (n = 10,731) collected during annual monitoring surveys at 21 sites throughout the upper Savage River watershed (2006-2014). Vertical dashed lines delineate the four stages used for simulation modeling in this manuscript**.**

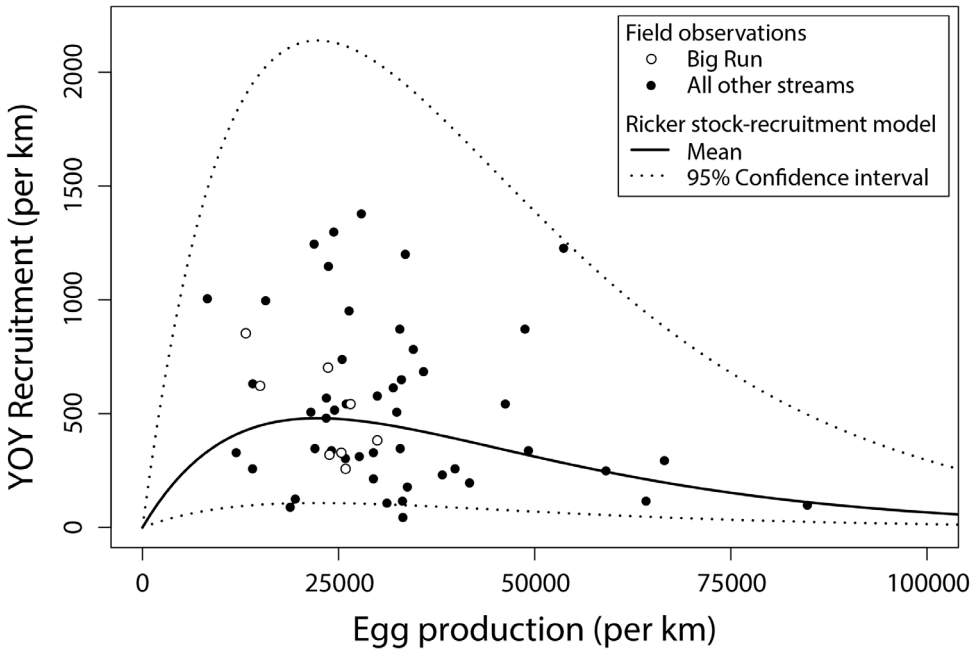

Figure 24. There was a weak (R^2 = 0.043) but statistically significant stock-recruitment relationship observed in a network of 21 sites on tributaries to the Savage River (2006-2014; $P < 0.05$). Egg production estimates assume a 1:1 sex ratio and are based on the length-fecundity relationship reported by Letcher et al. (2007). Young-of-the-year densities were based on annual monitoring surveys, and assume a 50% capture probability. Hollow symbols represent surveys conducted on Big Run; solid symbols represent other survey locations.

Scenario Results

Under the base scenario, populations were not at risk of extinction within 50 years (0%). Adult abundance varied considerably (606-6,252 adults; mean = 3,773). These patterns are largely due to the pulse-driven nature of recruitment (309-10,563 recruits per year; mean = 6,958). Under these conditions, the average generation time was estimated to be 2.97 years. Mean annual per capita recruitment was 1.27 recruits per small female; 2.63 recruits per medium female; and 4.564 recruits per large female. Large individuals generally comprised a small component of the adult population (median = 25.5%), but were responsible for over half of total egg production in 26.2% of years, highlighting their role in sustaining the population through down years. Approximately half of the populations (49.1%) returned to the recovery target (3,773 adult females) within ten years following a severe population crash (two adult females per 100 m of stream).

Growth regime shifts due to environmental change altered Brook Trout size structure in Big Run. When fast growth years were more common, more Brook Trout reached larger sizes (Figure 25). Growth patterns also had a small but significant impact on the rate of population recovery (1.0% of variance explained; $P < 0.001$; Figure 26). Mean generation times were not strongly influenced by growth patterns (Table 17).

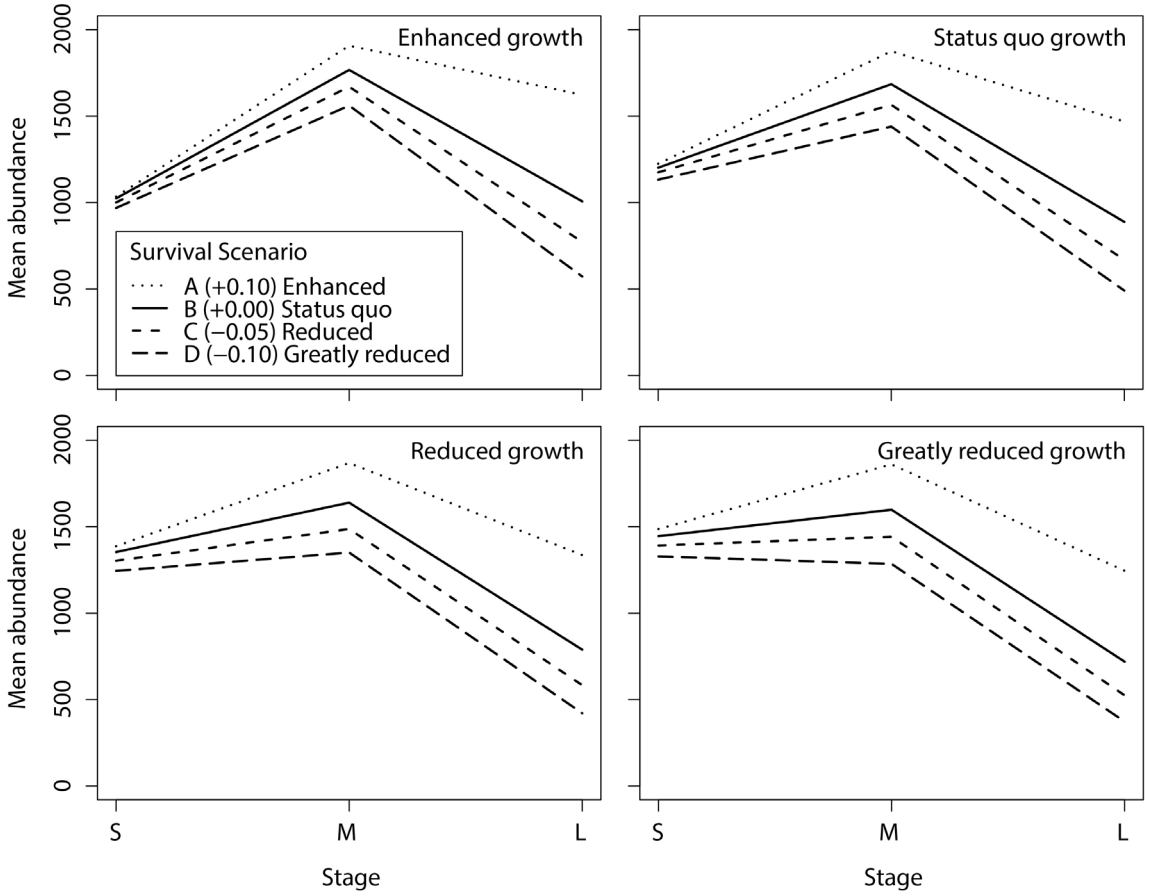

Figure 255. Mean size structure of simulated Brook Trout populations under four different growth regimes. Each line represents a specific survival scenario and is distinguished by a unique dash pattern.

Table 17. Mean generation time for simulated Brook Trout populations under current and alternative scenarios.

Survival	Growth			
	1	2	3	4
A	3.38	3.40	3.42	3.43
B	2.96	2.97	2.98	2.99
C	2.79	2.80	2.81	2.82
D	2.64	2.57	2.65	2.66

Changes in adult survival influenced the resilience and size structure of Brook Trout populations. Increased adult survival had a significant positive effect on population resilience ($P < 0.001$; 16.3% of variance explained), as measured by the rate of recovery following a severe population crash (Figure 26). When adult survival rates were increased by 10%, populations were significantly more likely to reach their recovery target within 10 years when compared with populations with unchanged survival rates (70.7% versus 49.1%). Greater adult survival also resulted in a marked increased abundance of large fish (Figure 25). Conversely, a 10% reduction in adult survival from the observed patterns significantly reduced the likelihood a population would recover within a decade (25.8% versus 49.1%). Far fewer large individuals were represented in populations with reduced and greatly reduced survival than under the status quo scenario. Mean generation times were somewhat longer for simulations with higher survival rates (Table 17). A 30% reduction in annual recruitment dramatically reduced the likelihood of population recovery within a decade when compared to the base case (12.8% versus 49.1% ten years post-disturbance). None of the simulated populations in any of our growth and survival scenarios were projected to be extinct within 50 years.

The average relative reproductive value for each stage also varied across the different scenarios (Figure 27). Under conditions of decreased growth and survival, large Brook Trout are the most valuable with medium-sized individuals also of enhanced value. The relative reproductive value of small Brook Trout was stable across the scenarios, but marginally higher under conditions of high survival and rapid growth.

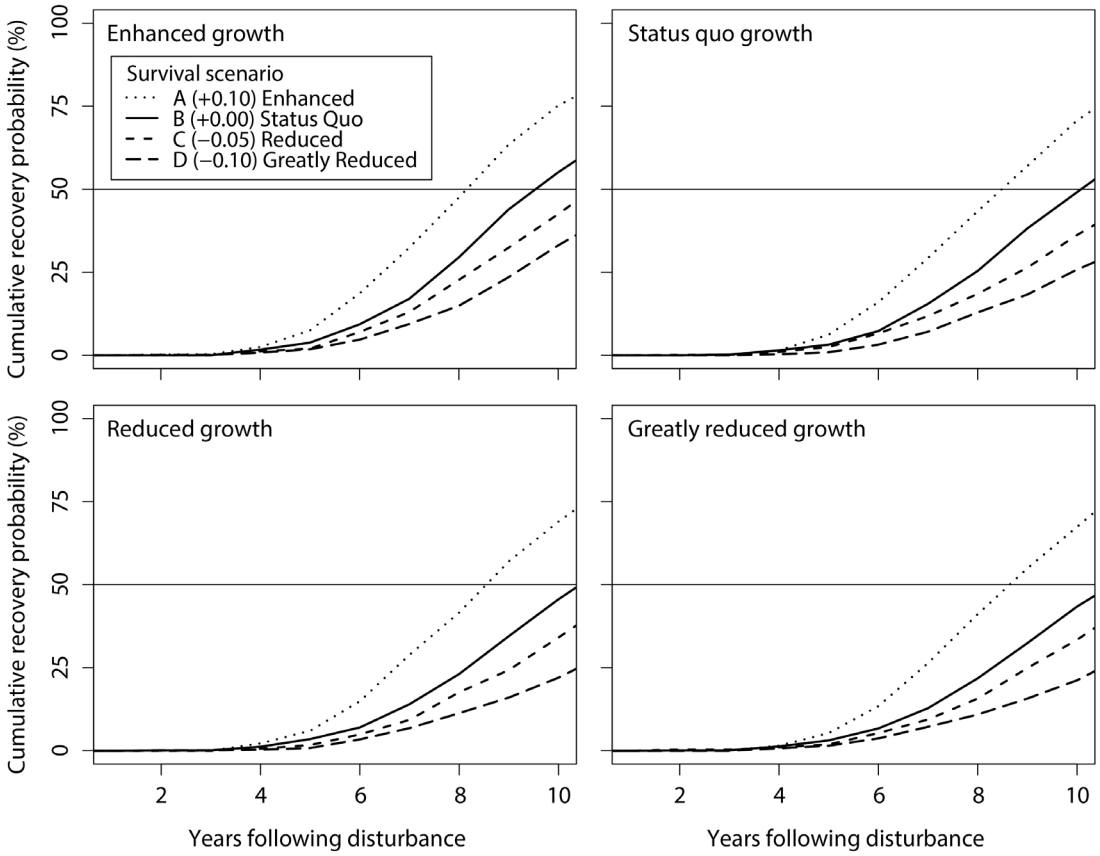

Figure 26. Cumulative probability of recovery for simulated Brook Trout populations under four different growth regimes. Each line represents a specific survival scenario and is distinguished by a unique dash pattern.

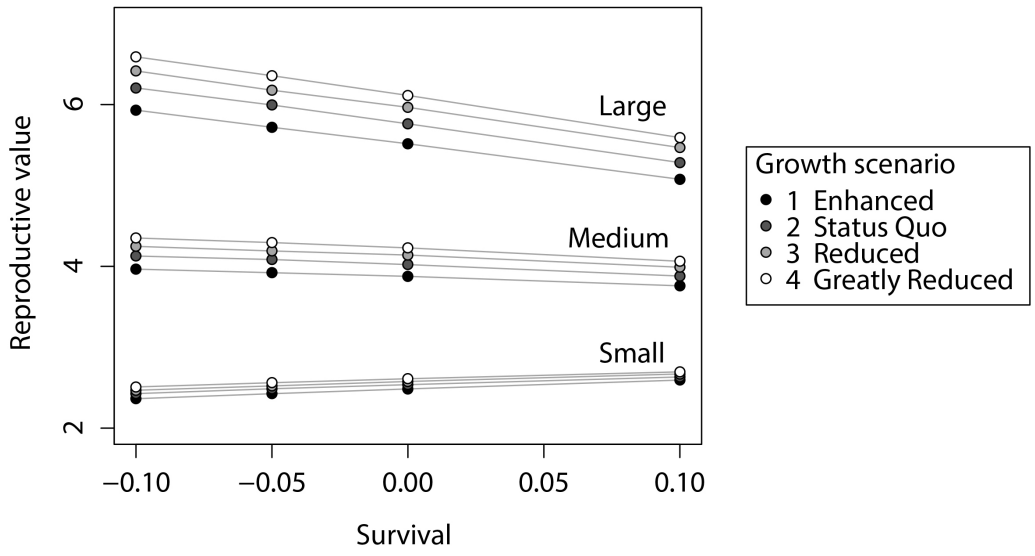

Figure 27. A comparison of the reproductive value of small, medium, and large Brook Trout under an array of different survival and growth scenarios.

Discussion

Population dynamics were dominated by pulse-driven recruitment under all scenarios. Small numbers of spawners were often capable of producing high recruitment. While the result follows directly from our model structure, the field data showed no relationship between total fecundity and recruitment. This results in erratic patterns of abundance and has been observed in other salmonid species and different regions (e.g., Platts and Nelson 1988). In fact, many populations of exploited fishes lack stock-recruitment relationships across normal ranges of abundance, and environmental controls frequently drive recruitment patterns (Szuwalski et al. 2014). Consequently, population growth rates (λ) ranged widely (mean = 1.09; range = 0.35-5.04). Whereas adult survival varied across a moderate range ((maximum relative fluctuation = 1.41; 32.9-45.8%) in our base scenario, recruitment variability was much greater, thus illustrating the sensitivity and influence of recruitment variability on the future population.

Environmental conditions can vary considerably from year to year to influence not only population dynamics, but even community structure (Zalewski and Naiman 1985). Previous studies have reported that floods (Needham and Jones 1959, Seegrist and Gard 1972, Hoopes 1975), water chemistry (Trojnar 1977, Cleveland et al. 1986, Ingersoll et al. 1990), temperature and ice conditions (Latta 1968, Hunt 1969, Curry et al. 1994) can have strong influences on the survival of young Brook Trout. Latta (1965) found no relationship between spawners and recruits, but reported a strong relationship between groundwater levels and recruitment. Although we did not specifically examine environmental variables, we infer that environmental stochasticity dominates Brook Trout recruitment in tributaries to the upper Savage River.

Pulse-driven recruitment is a critical assumption in our model. Although the monitoring data supports this assumption, our results will be influenced if it is incorrect. Young-of-the-year catchability affects the magnitude of our modeled recruitment pulses, but catchability can vary widely based on environmental conditions and fish size (Borgstrom and Skaala 1993, Kruse et al. 1998, Niemelä et al. 2000). Given that we surveyed small headwater streams with relatively simple habitat, we believe our estimates here are also conservative, and parameterizing the model with higher assumed catchability results in both lower population abundances and resilience. For example, using the actual numbers of captured young-of-the-year rather than adjusting for catchability resulted in only 12% of populations achieving the recovery target within four years of a disturbance.

Unlike many species whose population attributes buffer against recruitment failures with long adult life spans, Brook Trout are relatively short lived and rely on frequent pulses of recruitment to sustain their populations in headwater streams such as the Savage River watershed. Management and restoration activities that promote environmental stability and thus stabilize recruitment patterns may help to promote population resilience. Yet that same sensitivity also suggests that the predicted increases in environmental variability coincident with global climate change may have disastrous consequences for the resilience and even persistence of wild Brook Trout. Should the frequency of below average recruitment increase without management actions increasing longevity or adult abundances, many populations may not recover sufficiently before the next recruitment pulse. Such populations will continue to persist over the short-term, but will slowly spiral to extirpation (e.g., Hilderbrand 2003). Even though we did not observe

any simulated extirpations, the sensitivity of the system to key parameters coupled with the potential for an extinction debt (Kuussaari et al. 2009) that takes time to manifest highlight the vulnerability of the stream-dwelling Brook Trout populations. Future work should examine the influence of changing patterns of recruitment on population dynamics and consider how managers may be able to enhance and stabilize the recruitment of young Brook Trout.

Contrary to initial expectations, our range of growth scenarios had only a small impact on the resilience of Brook Trout populations. Since egg production was only weakly related to recruitment (Figure 24), growth has a limited impact on the population's reproductive potential and primarily serves to increase an individual's size. It is possible that growth (and thus body size) confers survival benefits (Hutchings 2006). However, using our dataset we were unable to generate meaningful estimates of survival for all four stages represented in our projection model. Consequently, we assumed all adult fish (≥100 mm) survived at the same rate. Regardless, unless there are marked differences in survival among size groups of adult Brook Trout, we do not expect changes in growth rate to have a strong direct influence on population resilience.

Changes in adult survival had a considerable impact on the resilience of simulated Brook Trout populations. Enhanced adult survival allowed the population to retain a greater number of recruits, thus attaining recovery targets in less time. A 10% increase in adult survival from the status quo reduced the median recovery time by nearly two years - which is likely to be very important in a system where the mean generation time is roughly three years. Given that less favorable environmental conditions are projected to become more common (US EPA 2001, IPCC 2001), it is reasonable to expect that years

with poor recruitment or low survival will become more common. Under these circumstances, faster recovery times will promote population persistence.

Pronounced impacts of changing adult survival were also evident in the size structure and abundance of the simulated populations. Enhanced adult survival reduced the rate of population decline between good recruitment years, and yielded a higher average abundance of adults with more large fish, thus improving the quality of the fishery. In general, we expect the impact of changes in survival on population dynamics to be more important in longer lived populations and management to promote resilience may be more effective in these cases. Across all simulations, however, pulse-driven recruitment patterns commonly produced large swings of adult abundance.

Our study suggests fishing regulations may have some utility to promote population resilience in western Maryland. It also suggests that the aspects of Brook Trout ecology we examined are naturally somewhat resilient to environmental change. In general, *Salvelinus* spp. are well-adapted to dynamic environments (Power 2002), but are sensitive to warming conditions (Lehtonen 1998, Gunn and Snucins 2010, Robinson et al. 2010, Gerdeaux 2011). Brook Trout are probably far more susceptible to population collapse due to changing patterns of recruitment or periods of lethal stream temperatures than to relatively small changes in the vital rates of adults. Unfortunately, both of these scenarios are likely under current climate change projections, and warrant further attention.

Our findings and recommendations are specific to populations without a strong stock-recruitment relationship. Since the abundance of spawners and egg production is not related to recruitment, recruitment overfishing is unlikely to occur in our study

system. In populations where egg production is correlated with recruitment, we expect our results may be different. First, we expect changing growth regimes would have a stronger influence on population resilience. Growth and survival patterns have a considerable influence on the abundance of large, highly fecund individuals (Figure 25). These individuals are disproportionately important to recruitment. Additionally, these populations may be slower to recover and more vulnerable to extirpation, as reproductive output would be reduced during times of low abundance. Consequently, the utility of traditional tools to enhance the resilience and persistence of wild Brook Trout populations is likely greater where stronger stock-recruitment relationships exist.

Where environmental change significantly jeopardizes Brook Trout, regulatory measures alone are unlikely to be adequate to conserve wild populations. Based on our simulations, the greatest opportunity to mitigate against environmental changes relate to enhancing patterns of recruitment. Given our limited ability and resources to enhance recruitment, Brook Trout populations may be best protected by addressing drivers of environmental change directly.

Acknowledgements

We would like to thank the Maryland Department of Natural Resources for providing the financial, logistical, and data resources necessary to complete this research. In particular, we would like to express our gratitude to Alan Heft, Alan Klotz, and the Mt. Nebo fisheries crew for providing extensive support with electrofishing samples. Numerous volunteers and technicians, including Erin Deck, Ryan Cooper, Mark Colaw, and Andrew Heft, provided assistance with field sampling.

Chapter 6: Synthesis

My research has changed our understanding of the structure and function of Brook Trout populations in western Maryland. My findings indicate that Brook Trout exhibit remarkably fine population structures. Multiple populations may co-occur in a small watershed even in the absence of physical barriers to movement. These populations may even exhibit differences in vital rates. Males and females also exhibit marked differences in life history, particularly with respect to annual growth rates. Additionally, I documented considerable spatial and temporal variation in life history. Overall, I characterized Brook Trout populations in a small headwater system in western Maryland and added to our already complex understanding of the species as a whole.

These results have important implications for the conservation and management of Brook Trout. First, contiguous patches of habitat that are often used to delineate populations (Whitely et al. 2013) may not adequately capture the underlying biological structures and diversity. Cryptic population structures may be widespread but have gone largely undetected. Although attempts to formally understand the adaptive differences between genetic groups are in their infancy, recent studies indicate local adaptation is widespread and presumably important to the persistence of populations. Undetected populations can represent unique genetic groups with potentially important local adaptation and warrant conservation. Furthermore, this also suggests some of our populations may be much smaller and therefore more vulnerable than previously thought. Large interconnected watersheds should not be assumed to represent a single, robust population. When populations are extirpated, biodiversity is lost forever.

Unfortunately, it is not feasible to individually manage, or even document, all populations across the species range. However, a basic recognition of the underlying biological complexity found in Brook Trout populations may help identify management strategies to conserve them. The fundamental unit of management should be the populations, and these are not always spread throughout interconnected watersheds with suitable habitat. Reintroduction programs should exercise special caution when sourcing their fish, as even fish collected within the same watershed may not be of the same population. If this occurs, translocated fish may remain reproductively isolated even when occurring in sympatry (Richards et al. 2008). Efforts to restore connectivity need to recognize that population-specific movement patterns may exist that effectively isolate fish even in the absence of physical barriers to movement.

In general, my results are congruent with many other studies on Brook Trout, but further add to our understanding of the complexity of the species. Brook Trout are an extremely plastic species, with considerable variation among populations. Geographically proximate populations can have marked differences in life history (Kazyak et al. 2013). Range-wide or even regional generalizations may fail to capture important differences among populations, and these differences may have important implications for the way we manage wild fisheries. Prior to this study, it was expected that Brook Trout in the upper Savage River watershed grow rapidly and seldom live past three years of age. While this is partially true, it fails to reflect the tremendous amount of interannual variability in growth rates or consider the presence of very large, old individuals that were observed in the mark-recapture component of this dissertation. Consequently, management strategies that assume fast growth and rapid turnover may fail to protect

large, highly fecund individuals that play a disproportionate role in sustaining populations through periods of low recruitment.

The conclusions presented in this dissertation rely on a few fundamental assumptions. First, we assume that marked fish provide an unbiased representation of the population as a whole. Passive integrated transponder tags are widely used (Marvin 2012) and are generally reported to have a very small influence on the growth (Ombredane et al. 1998, Ostrand et al. 2012) and survival (Dare 2003, Achord et al. 2007, Ostrand et al. 2012) of salmonids. Consequently, I do not expect tagging effects to have had a substantive influence on the results of the mark-recapture study.

For our simulation study, our model largely omits density-dependent processes. We assumed that there was a weak relationship between stock size or egg production and recruitment. Our field data supports this assumption, but the simulation model is quite sensitive to changing patterns of recruitment. In nearby streams in West Virginia, stock-recruitment relationships occur in some populations (Hartman et al. 2007). I anticipate populations with strong stock recruitment relationships would be more vulnerable to extirpation, resilience would be more directly linked to abundance, and large fish would be even more important to population dynamics. Furthermore, we may have failed to capture important linkages between abundance and growth or survival. The length of our study (3 years) precluded a robust examination of density-dependent effects on vital rates. Some studies have reported reduced growth (Jenkins Jr. et al. 1999, Utz and Hartman 2009, Grant and Imre 2005) or survival (Einum and Nislow 2005, Vøllestad and Olsen 2008) when salmonid abundance is high. Under these circumstances, density-dependent effects are expected to stabilize population dynamics (Murdoch 1994).

If I were to start the project over again, with an ability to foresee the future, I would make several changes. First, I would have restricted the study to the Big Run watershed. Although our mark-recapture work in Middle Fork provided an interesting comparison with Big Run in the growth study, the small spatial extent of our study area (0.5 km) resulted in comparatively few recaptured fish that could be used for subsequent analyses. In retrospect, this effort may have been better spent in the headwaters of Big Run. Since we only sampled the lowermost 4.5 km of the Big Run watershed, it would be interesting to see if vital rates were different at upstream locations. Additionally, it would have been very interesting to see if there were distinct genetic groups in Whiskey Hollow and on Miller Run, as an extension of Chapter 4.

In retrospect, I also would have positioned the PIT antennas differently. Rather than locating an antenna array at the upper limit of the study area on Big Run, I would have positioned it just upstream of the confluence of Monroe Run. This would have allowed a better estimation of the amount and timing of physical movement between the two genetically distinct populations observed within the study area. Also, if it were feasible, an antenna on the mouth of Big Run also may have been very enlightening. By chance, I located a single tagged fish from Big Run in the mainstem upper Savage River during the winter of 2011. Hence, we qualitatively know that Brook Trout from Big Run use the mainstem upper Savage River, but we have no idea of the prevalence of this behavior.

It would have been valuable to have collected standardized photographic vouchers for all individuals. This would have allowed assignment of sex (unpublished data) for all individuals, thus facilitating sex-specific estimates of survival. In addition, photographic

vouchers would have allowed a comparison of morphology between genetic groups. This would have been especially interesting in the lower reaches of Big Run, where fish from both genetic groups may co-occur. Finally, this data would have been a very interesting resource to study ontogenetic shifts in morphology through time. It may have even been possible to determine if certain morphotypes expressed different vital rates.

The research presented here would also have been enhanced by a longer time series of mark-recapture data. We characterized Brook Trout population dynamics during three years. The first winter of the study produced record snowfall followed by rapid melting and extreme flows. During the second year of the study, protracted periods of spring-like conditions appeared to have produced exceptional growth rates in our study population. It is unknown how frequently these conditions may occur or how well our three years of study represent long-term patterns of environmental stochasticity.

There are many important yet unanswered questions regarding the conservation and management of Brook Trout in western Maryland. For example, how much adult mortality is attributable to fishing? How much of an impact to the population dynamics, resilience, and persistence can be expected? The answers to these questions will help guide management decisions, particularly with regards to how to regulate angling. How much thermal refugia is available to mitigate against the predicted effects of climate change? An assessment of thermal sensitivity and local refugia will help us to predict the impacts of climate change on local Brook Trout populations. What strategies can be implemented to stabilize and improve Brook Trout recruitment? Little is known as to what strategies, if any, will be effective in enhancing wild Brook Trout populations in the upper Savage River watershed. To what extent do very small tributaries and disjunct

floodplain habitats contribute to the production of Brook Trout in headwater systems? We observed young of the year fish in both locations, but it is still unknown the extent to which these fish represent a viable contribution to the population, and how their contribution may change in the context of environmental change. Where multiple populations occur, do they possess unique local adaptations, or are these populations effectively demographically isolated duplicates of one another? Recent advances in genomics are making this question tractable (Meier et al. 2014), where previously we could only speculate. Addressing these questions will further improve our understanding of the structure and function of wild Brook Trout populations, and aid in the design of effective management strategies in a changing world.

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